UDK 630*453 (001)

Izvorni znanstveni članci – Original scientific papers
Šumarski list, 7–8 (2013): 395–401

COMPARISON OF ALL SEASON AND STANDARD TYPE OF ECOLURE® DISPENSER EFFICACY IN TRAP CATCHES OF EUROPAEAN SPRUCE BARK BEETLE (*Ips typographus* (L.))

USPOREDBA UČINKOVITOSTI CIJELOSEZONSKOG I STANDARDNOG FEROMONA ECOLURE® U ULOVIMA SMREKINOG PISARA (*Ips typographus* (L.))

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Summary:

I. typographus is the most serious pest of spruce forests in Eurasia. Pheromone traps are usually used in forest protection against this pest. In this study, two types of pheromone dispensers (ECOLURE CLASSIC and ECOLURE MEGA) were compared in terms of its efficiency in 2010. ECOLURE CLASSIC were capturing averagely more beetles in compare of all season dispense ECOLURE MEGA during all season. No statistic difference was recorded only during first 10 days of the season. In the rest of season (next 123 days) the ECOLURE CLASSIC captured statistically more beetles then ECOLURE MEGA. That is why, type and quality of pheromone dispenser significantly influences the number of trapped beetles to the pheromone traps.

KEY WORDS: efficiency, pheromone dispenser, ECOLURE, *Ips typographus*

Introduction
Uvod

The spruce bark beetle *Ips typographus* is the most damaging insect attacking spruce forests (*Picea abies* (L., 1753) Karsten, 1881) in Eurasia (Christiansen and Bakke, 1988). With regard to forest protection, the last two decades in Central Europe was marked by the storms "Vivian/Wiebke" in February/March 1990, "Lothar" in December 1999 and "Kyrill" in January 2007. All events were disastrous and gave rise to an enormous propagation of the European spruce bark beetle (*Ips typographus* /L./) in the affected spruce forests (Engesser et al., 2002; Flot et al., 2002; Schröter et al., 2002; Berec et al.,2013).

Adult beetles of *I. typographus* emerge from the forest litter and tree bark on warm spring days and fly to stressed host trees. These bark beetles use an aggregation pheromone to attract more individuals of the same species to the tree for the purpose of weakening the tree and mating. The pheromone attracts both sexes. The attracted males join the attack

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and secure an area for mating and oviposition. This area consists of a hole and a chamber beneath the bark known as a "nuptial chamber". The females construct a tunnel ("maternal gallery") beneath the nuptial chamber in which to lay eggs. In all species of the *Ips* genus, several females (usually two or three) join each male in his nuptial chamber (Wermelinger, 2004). At lower elevations, overwintering *I. typographus* beetles emerge in April/May and produce one or two generations of offspring per year with the main peaks of bark beetle emergence in July and August/September. At elevations above 1,000 m a.s.l., this species produces only one generation of offspring per year (Wermelinger, 2004). Trap trees using and removing of infested logs from the forests are basic tools of the forest protection against *Ips typhraphus* during these days. Also pheromone traps are massively used for these purposes in spite of their still discussed efficiency by many authors (e.g Dimitri et al., 1992; Lobinger and Skatulla, 1996; Wichmann and Ravn, 2001). Some authors show that only 3–10 % of bark-beetle population may be trapped by using of high density of the pheromone traps (Weslien and Lindelöw, 1990; Lobinger and Skatulla, 1996). Their potential efficiency can be furthermore decrease by capturing of predators of the beetles. Some special construction of pheromone traps was suggested for reducing of this negative effect (Martin et al., 2013) for related species of *I. sexdentatus* (Börner, 1776). Also type of wrapper play very significant role in number of trapped beetle (Nakládal and Sova, 2010). Bakke et al. (1989) discovered that pheromone dispensers containing methylbutenol, (–) cis-verbenol and ipsdienol, which are the three components of the *Ips typographus* pheromone, retained their effectiveness for 8 years when they are stored in sealed aluminium bags in an unheated room in the cellar of a storehouse.

A key component of the lures for pheromone traps is cis-verbenol (e.g. Jakuš and Blaženec, 2002). Many types of pheromone dispensers (based on this component) with different efficiency are in the market during these days. E.g., Zahradník et al. (1990) compared efficiency of PHEROPRAX and IT ETOKAP. Holuša et al. (2010) compared efficiency of ID Ecolure, Pheagr IDU and Duplodor for the double spined bark beetle (*Ips duplicatus* /C.R. Sahlberg, 1836/). Jakuš and Šimko (2000) compared IT ECOLURE (with 6 different levels of release rates) and PHEROPRAX at pheromone trap barriers.

In last time, the all season pheromone dispensers begin offered in the market. Eg., guaranteed efficiency of 18–20 weeks is declared for both ECOLURE TUBUS and ECOLURE MEGA. These dispensers do not require additional opening dispenser bag and also their replacement. This strong advantage may be counterbalanced by their obvious low efficiency (Nakládal and Sova, 2010). Basic difference between classic ECOLURE CLASSIC and all season ECOLURE TUBUS dispenser is only in type of releasing wrapper. Effective compounds are packed in classic clipping bag in case of ECOLURE CLASSIC, and in special gauzy plastic tube with free filling in case of ECOLURE TUBUS. ECOLURE MEGA tested in this experiment has similar construction like ECOLURE CLASSIC.

The aim of this study is to compare the efficiency of ECOLURE MEGA (all season dispenser) and efficiency of ECOLURE CLASSIC (standard dispenser).

Material and methods
Materijali i metode

The efficiency of the standard (ECOLURE CLASSIC) and all season (ECOLURE MEGA) pheromone dispensers was evaluated in this experiment in 2010. ECOLURE MEGA has guaranteed efficiency of 18–20 weeks. This time was taken like comparing of efficiency of the ECLURE CLASSIC (with efficiency of 5 weeks after first clip of wrapping bag, second clip prolong efficiency by next 7–10 days). IT ECOLURE CLASSIC (rank 080411) and IT ECOLURE MEGA (rank 080411) were used for the test. Both are pheromone dispensers on *Ips typographus* compounding from (S)-cis-verbenol (3 %), alcohols and solvents (85.2 %) and synergic components (11.8 %). ECOLURE CLASSIC has 2.5 g of effective compounds and ECOLURE TUBUS has 3 g in comparison.

The study was made in commercial forests near the Písek city (south Bohemia – Czech Republic) in Záhoří management-plan area sited near the Záhoří village (Loc: 49°21'1"N, 14°12'1"E). Twenty pairs of Theysohn type of pheromone traps (one trap in pair with ECOLURE MEGA and one with ECOLURE CLASSIC dispenser) were installed to the forest complex with average age of 90 year and 70 % of spruce portion of surrounding forests. Exact data and position of all pairs of traps are mentioned in table 1.

Pheromone traps were positioned on clear-cuts in 15 m distances from the forest edge (in according with commending of pheromone dispenser producer). Distance of traps in pair was 70 m by following way that both were placed on stand wall oriented on the same cardinal point. Strongly weedy places were measured by herbicides 1.5 m around the trap.

The comparing duration of experiment was 133 days in 2010. The experiment was finish in guaranteed efficiency term of ECOLURE MEGA (18–20 weeks = 126–140 days). Pheromone traps were lured at 1st May by pheromone dispensers. Traps were checked every 10 days approximately to the 10th September (exact date of sampling: 10th May, 20th May, 30th May, 12th June, 22nd June, 2nd July, 12th July, 22nd July, 1st August, 11th August, 21st August, 31st August, 10th September).

The first bag with efficiency substance of ECOLURE CLASSIC was more opened by scissors in 5th June and replaced by second one in 12th July. The second was more opened 17th June and replaced by third one in 22nd July. The third

Table 1. Position parameters of centers all pairs of traps, stand no, spruce portion (%) and age of surrounding stand.
Tablica 1. GPS koordinata između klopki, ime sastojine, dob sastojine, udio smreke (%) u sastojini.

pair of traps no. parovi klopki	stand no. pozicija	age starost	Spruce portion in stand (%) udio smreke u sastojini (%)	GPS position of traps (centre between them) GPS koordinata klopki (središte između klopki)
1	951 A 11	105	85	49°23'4.461"N; 14°10'41.623"E
2	951 D 9	89	80	49°23'4.604"N; 14°10'38.992"E
3	951 D 9	89	80	49°23'1.095"N; 14°10'28.337"E
4	951 B 10	100	80	49°22'56.441"N; 14°10'11.352"E
5	951 E 11	105	80	49°22'56.878"N; 14°10'27.316"E
6	952 B 5	50	50	49°22'2.69"N; 14°11'34.321"E
7	952 F 6	52	65	48°21'28.259"N; 14°11'23.879"E
8	952 G 6	52	70	49°21'26.798"N; 14°11'32.628"E
9	952 G 6	52	70	49°21'23.214"N; 14°11'34.372"E
10	953 E 11	108	80	49°20'53.196"N; 14°11'38.778"E
11	953 E 11	108	80	49°20'54.114"N; 14°11'35.988"E
12	953 D 9	87	50	49°21'2.435"N; 14°11'30.994"E
13	952 D 13	130	45	49°21'41.272"N; 14°11'23.12"E
14	954 E 11	110	90	49°22'21.087"N; 14°14'31.851"E
15	954 G 6	55	60	49°22'8.318"N; 14°14'45.139"E
16	954 F 6	55	55	49°22'17.632"N; 14°14'28.483"E
17	954 A 11	110	85	49°22'39.012"N; 14°14'59.349"E
18	954 B 10	98	95	49°22'27.623"N; 14°15'10.879"E
19	953 A 14	137	56	49°21'17.94"N; 14°11'39.366"E
20	955 C 10	98	90	49°20'9.828"N; 14°15'27.385"E

one was more opened in 26th August and replaced by fourth in 31st August which was not more opened late. Number of trapped beetles was counted in accuracy of 1 beetle. In case of numerous samples (more the 35 beetles), the calibrated glass cylinder was used for this purpose (1 ml of beetles = 35 specimens of *I. typographus*).

Software STATISTICA 9.1 was utilized to the data analysis. The following procedure was made repeatedly for each check. At first, the differences of trapped beetles (ECOLURE CLASSIC – ECOLURE MEGA) in each pair of traps were calculated. The normality of the differences was tested by Shapiro-Wilk W test. Convenient transformation function was applied on origin data to remove non-normality from the differences. *T*-test for dependent samples was used on such data. Wilcoxon matched pairs test was applied on data where didn't find convenient transformation function.

Relative efficiency was calculated for each checking like rate of number trapped beetles to the trap lured by ECOLURE CLASSIC divided by number of beetles trapped to the trap lured by ECOLURE MEGA (C/M index).

All trapped beetles were continuously fixed in ethyl-alcohol in one container for each type of dispenser separately during all season. One hundred beetles from each container were sampled to examination of male portion. Sexes were determined with use to dissection of genitals. Character of obtained data doesn't allow statistic testing of this part of research.

Results
Rezultati

Prikupljeni podaci

During the whole season 179,477 beetle were trapped to the all 40 pheromone traps. 32 % of all were captured to the pheromone traps lured by ECOLURE MEGA and 68 % to the traps lured by ECOLURE CLASSIC. It means that ECOLURE CLASSIC was 2.1 times more effective then ECOLURE MEGA.

Total numbers of trapped beetles for each checking and both types of dispensers is presented on Fig. 1. The figure show usual course of number trapped beetles. There are two obvious peaks representing peaks of spring (May 20th) and of summer (July 12th) swarming.

Usporedba učinkovitosti

The traps lured by ECOLURE CLASSIC captured always more count of beetles than ECOLURE MEGA (from 1.1 to 112.4 times more – see Fig. 1 on C/M index). Differences

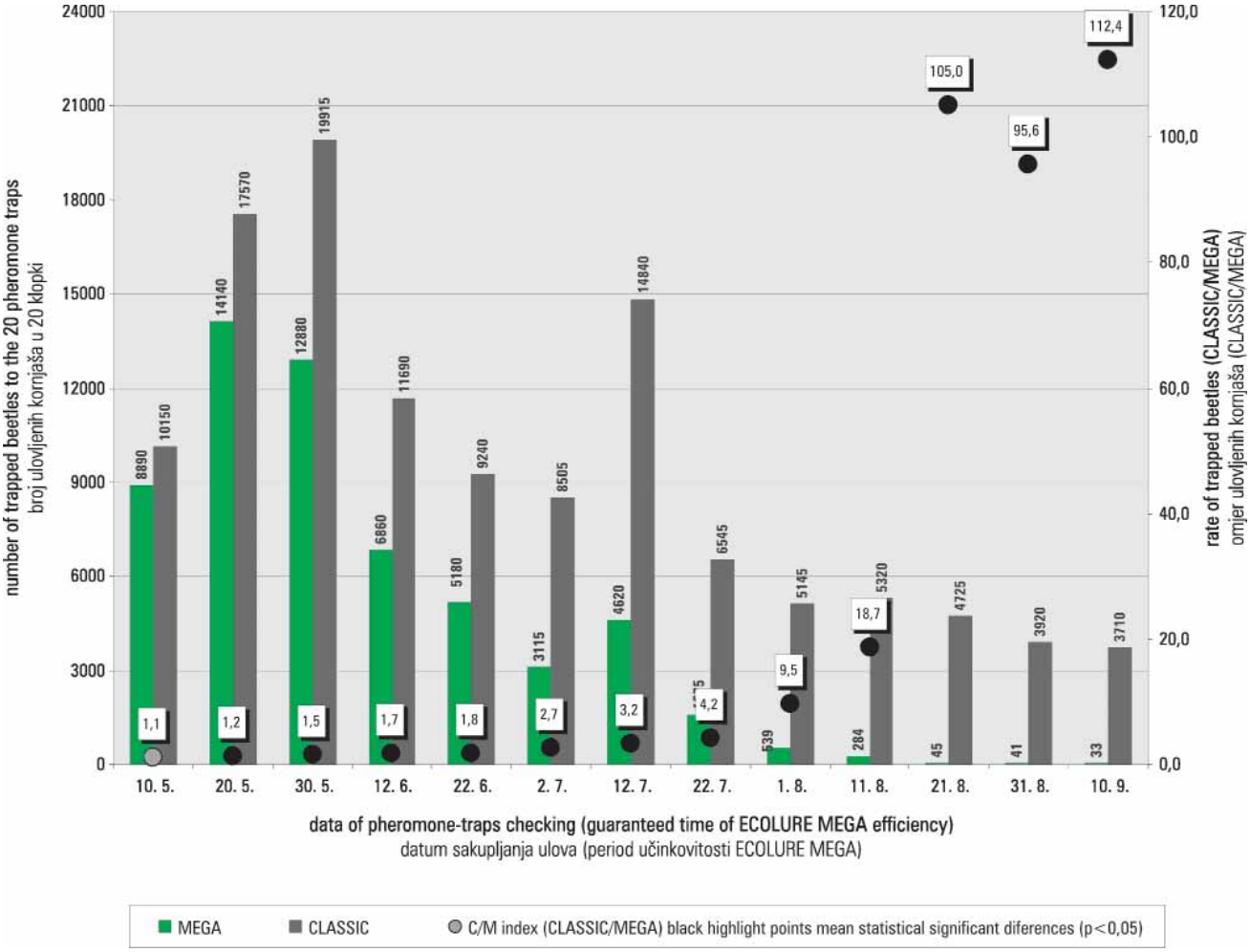

Figure 1. Trapped beetles results with ECOLURE CLASSIC and ECOLURE MEGA dispensers during the season of beetle flight activity. Guaranteed effective duration of ECOLURE MEGA represents X axis (18–20 weeks) (19 weeks are on X axis in fig. 1).

Slika 1. Rezultati ulova kornjaša na ECOLURE CLASIC i ECOLURE MEGA raspršivačima tijekom perioda praćenja. Jamčena učinkovitost ECOLURE MEGA raspršivača predstavljena je osi X (18–20 tjedana) (19 tjedana je na osi X u Fig.1).

between number of trapped beetles were statistically significant except for only the first checking ($\alpha = 0.05$) – see Tab. 2. During spring swarming (checking May 10th to July 2nd) ECOLURE CLASSIC trapped 1.1–2.7 times more. The relative efficiency of ECOLURE CLASSIC continuously weakly grew in this period.

In contrary, relative efficiency swiftly very grew during second swarming (July 2nd to October 10th). ECOLURE CLASSIC trapped from 2.7 to 112.4 times more beetles in compare with ECOLURE MEGA.

Convenient transformation function for satisfy to parametric test assumption and statistical evaluations including p-values for all checks are presented in Tab. 2.

Istraživanje omjera spolova

The efficacy from this point of view was the same for both of dispensers' type. In both samples were recorded 92 female and 8 males. This data was not tested due to missing differences between these results.

Discussion
Rasprava

Two obvious generations per year was recorded during the survey season in this paper alike as recorded Nakládal and Sova (2010). This is common in Central Europe, except at higher elevations (Wermelinger and Seifert, 1999).

Overall, the results show that all season ECOLURE MEGA dispenser is not suitable in comparing with ECOLURE CLASSIC in common forestry conditions. These result strictly corresponded with conclusions published by Nakládal and Sova (2010), which tested ECOLURE CLASSIC in comparison to other type of all season pheromone dispenser ECOLURE TUBUS. In both of these studies, ECOLURE CLASSIC trapped always more beetles then all season types dispensers during the trapping season. The relative efficiency of ECOLURE CLASSIC went slowly up (moving from 1.1 to 2.7) during the spring swarming. Nakládal and Sova (2010) recorded rapid shrinkage of all season dispenser efficiency during second swarming (moving from

Table 2. Statistical analysis for each sample. TT – *t*-test for dependent samples, WT – Wilcoxon matched pairs test; W – Shapiro-Wilk W test. P-values lower the 0.05 are highlighted.

Tablica 2. Podaci statističke obrade za svaki uzorak. TT – t-test za ovisne uzorke, WT – Wilcoxon test sparenih uzoraka; W – Shapiro-Wilk W test. P – vrijednosti manje od 0.05 označene su masnim tiskom.

Date of checking datum sakupljanja ulova	Transformational function (x_t) transformacijska funkcija (x_t)	normality (W test) *p*-values normala (W test) p-vrijednost	Statistical diferences (type of test) statistička razlika (tip testa)	Statistical diferences *p*-values statistička razlika p-vrijednost
10th May	$x_t=x$	0.018977	WT	0.083231
20th May	$x_t=x$	0.000359	WT	0.006634
30th May	$x_t=x$	0.000222	WT	0.000089
12th June	$x_t=\log(x)$	0.310844	TT	0.000000
22nd June	$x_t=x$	0.531707	TT	0.000001
2nd July	$x_t=x$	0.031095	WT	0.000089
12th July	$x_t=x$	0.61970	TT	0.000000
22nd July	$x_t=x^2$	0.701537	TT	0.000000
1st August	$x_t=x$	0.762556	TT	0.000000
11th August	$x_t=x$	0.861196	TT	0.000000
21st August	$x_t=x$	0.138660	TT	0.000000
31st August	$x_t=\log(x+1)$	0.918438	TT	0.000000
10th September	$x_t=x$	0.664624	TT	0.000000

2.7 to 36.6 times lower). In the present study, the rapid efficiency shrinkage of all season dispenser was much more progressive (moving from 2.7 to 112.4 times lower). That is why ECOLURE MEGA should be considered as comparably less efficient type of all season dispenser then ECOLURE TUBUS.

Number of trapped beetles strongly depend on many environmental factors and local conditions, such as temperature, exposition, sun exposure and others (Lobinger, 1995). We highgly eliminated influence of these factors during the study by type of used statistic test. Furthermore, the position of all traps was exactly the same as used Nakládal and Sova (2010) in 2008. But all 20 traps lured by the standard dispenser (ECOLURE CLASSIC) by Nakládal and Sova (2010) in 2008 were lured by the all season dispenser (ECOLURE MEGA) in this experiment. And vice versa, all 20 traps lured by the all season dispenser (ECOLURE TUBUS) in 2008 were lured by the standard dispenser (ECOLURE CLASSIC) in this experiment. This methodology supported correctness of data interpretation of both studies and show that all season dispensers are much less effective then standard dispensers.

Low number of trapped beetles closely corresponds with lower pheromone released level to the environment (and it immediately after the season beginning). This fact relate with higher portion of males in trapped sample and others problems reviewed by Nakládal and Sova (2010).

Interesting is very low average portion of male (8 % only) for both of type of dispensers. Many authors discuss factors influenced male portion in catches. The sex ratio of beetles caught can vary according to the trap type (Bakke et al., 1983), different sex behavior during the tree colonization (Byers, 1983), pheromone mixture and its different release level of (4S)-cis-verbenol (Jakuš and Blaženec 2002). Portion of male may be influenced also by present males in environment. Although some authors indicate a percentage of *I. typographus* males after egg hatching of about 50 % (Merker and Wild 1954; Annila, 1969) and sex ratio of emerging beetles 1:1 (Botterweg, 1982), some study show population density depending sex ratio (Annila, 1971; Lobinger, 1996). Portion of captured males in pheromone traps usually vary about 35 %. Botterweg (1982) indicate 30.6 %, Zumr (1982) 39 %, Faccoli and Buffo (2004) 33.1 % respectively 37.7 % in two years experiment. Eight percent, recorded in this study, is extremely low value which is not possible to satisfactorily explain.

Conclusion
Zaključak

Efficiencies of standard (ECOLURE CLASSIC) and all season dispensers (ECOLURE MEGA) were compared. Both dispensers contains the same chemical components, on the other hand they have different packs. All season dispensers (ECOLURE MEGA) trapping less beetles in compare of ECOLURE CLASSIC immediately after their setting dispensers in the forest.

Statistical difference of number trapped beetles was non significant during only the first checking (only first 10 days) and then was highly significant (next 123 days).

Generally, ECOLURE all season dispensers trapping lower count of beetles in compare of classic dispenser (ECOLURE CLASSIC). ECOLURE all season dispenser may be usable in inaccessible terrains (etc.) where we assume checking period exceed 50 days. They are weakly effective 60 days after the setting on and practically non-effective later. The ECOLURE all season dispensers have from 36.6 to more over 100 times lower recorded efficiency in the end of declared efficiency in compare with ECOLURE CLASSIC.

Acknowledgements
Zahvala

This paper was written in the framework of the research project CIGA No. 20124310 (Czech University of Life Sciences Prague) "Prognostic model for identification of nun moth (*Lymantria monacha* /L./) outbreaks in expected climatic changes in the Czech Republic". We are also obliged to Jaroslav Sova (Svatonice, Czech Republic).

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Sažetak:

U današnje vrijeme glavni način zaštite od smrekinog pisara *Ips typographus* L. je postavljanje lovnih stabala i njihovo pravovremeno uklanjanje iz šume. Isto tako feromonske klopke često se koriste kao sredstvo zašite unatoč njihovoj dvojbenoj učinkovitosti (npr. Dimitri et al. 1992; Lobinger, Skatulla 1996; Wichmann, Ravn 2001). Neki su autori pokazali da samo 3–10 % populacije potkornjaka može biti ulovljeno kod visokog broja korištenih feromonskih klopki (Weslien, Lindelöw 1990; Lobinger, Skatulla 1996). Ključna komponenta mamaca za smrekinog pisara je cis-verbenol (e.g. Jakuš, Blaženec 2002), a na današnjem tržištu prisutno je puno različitih tipova feromonskih raspršivača koji se baziraju na ovoj aktivnoj komponenti. U broju ulovljenih potkornjaka značajnu ulogu ima i oblik ampule feromonskog sredstva (Nakladal, Sova 2010). U posljednje vrijeme na tržištu je prisutno sve više cijelosezonskih feromonskih raspršivača sa zajamčenim periodom djelotvornosti od 18–20 tjedana za ECOLURE TUBUS i ECOLURE MEGA. Ovi raspršivači ne zahtijevaju dodatno mijenjanje ili otvaranje tijekom sezone lova. Ove prednosti raspršivača mogu biti umanjene slabim učinkom (Nakladal, Sova 2010). Temeljna razlika između ECOLURE CLASSIC I ECOLURE TUBUS raspršivača je u tipu otpuštajućeg omota. ECOLURE MEGA testiran u ovom istraživanju ima sličnu konstrukciju kao ECOLURE CLASSIC. U ovom istraživanju uspoređivani su feromonski raspršivači ECOLURE CLASSIC i cijelosezonski ECOLURE MEGA. ECOLURE MEGA ima zajamčenu učinkovitost od 18 do 20 tjedana. Istraživanje je provedeno na lokaciji Pisek južna Češka u području smrekove kulture Zahori (49°21'01"N, 14°12'01") u razdoblju od 1. svibnja 2010. godine do 10. rujna 2010. godine. Tijekom 133 lovna dana korišteno je 20 parova Theyson klopki, od kojih je jedna klopka unutar para bila kompletirana s ECOLURE CLASSIC raspršivačem, a jedna ECOLURE MEGA. Klopke su bile postavljene na sječinama 15 metara od ruba šume i udaljenosti 70 metara između klopki istog para. Korov oko klopki bio je tretiran herbicidima. Svakih deset dana vršeno je sakupljanje uzoraka iz klopki u navedenom razdoblju. Prebrojavanje potkornjaka vršeno je pojedinačno, a kod većih uzoraka korištena je volumetrijska metoda 1 mL=35 smrekinih pisara. U ulovima potkornjaka vršeno je i određivanje omjera spolova pomoću disekcije genitalija. Statistička obrada podataka izrađena je pomoću softwarea STATISTICA 9.1. S obzirom na dobivene ulove i distribucije podataka korišteni su Shapiro-Wilk W test, T-test i Wilcoxon test poklapajućih parova. Tijekom cijele lovne sezone ulovljeno je 179 477 potkornjaka u 40 feromonskih klopki, 32 % od tog ulova uhvaćeno je raspršivačem ECOLURE MEGA, a 68 % ECOLURE CLASSIC. Potpuni ulovi potkornjaka prikazani su na Fig. 1., a transformacijske funkcije koje zadovoljavaju parametar statističkih testova u Tab.2. Uspoređujući učinkovitost ulova u proljetnom dijelu rojenja (od 10. svibnja do 2.srpnja) ECOLURE CLASSIC imao je 1.1–2.7 puta više ulova. U periodu ljetnog rojenja (2.srpnja do 10.listopada) ECOLURE CLASSIC imao je od 2.7 do 112.4 puta više ulova od ECOLURE MEGA Tab 2. Kod omjera spolova učinkovitost oba tipa raspršivača bila je podjednaka 92 % ženki i 8 % mužjaka. Tijekom ovog istraživanja uočene su dvije generacije smrekinog pisara, što je normalno u središnjoj Europi osim na većim nadmorskim visinama (Wermelinger, Seifert 1999). Sveukupni zaključak koji proizlazi iz ovog istraživa je da cijelogodišnji raspršivač ECOLURE MEGA u usporedbi s raspršivačem ECOLURE CLASSIC nije pogodan za uporabu u uobičajenim uvjetima šumarstva. Slični rezultati dobiveni su u usporedbi cjelogodišnjeg raspršivača ECOLURE TUBUS i ECOLURE CLASSIC (Nakládal and Sova 2010.). U ovom istraživanju učinkovitost ECOLURE MEGA raspršivača je naglo padala (od 2.7 do 112.4 puta) u odnosu na ECOLURE CLASSIC, za razliku od usporedbe ECUOLURE TUBUS koji je tijekom drugog rojenja imao pad učinkovitosti (od 2.7 do 36.6 puta) u odnosu na ECOLURE CLASSIC (Nakládal and Sova 2010). To govori da je ECOLURE MEGA nešto lošiji tip raspršivača od ECOLURE TUBUS raspršivača. Moguće utjecaje na ulov zbog pozicije klopki isključili smo promjenom pozicije cjelogodišnjih raspršivača na poziciju kontrolnih klopki i obratno. Slabi ulovi u klopkama su u korelaciji sa slabim otpuštanjem feromona u okolinu, a to se uočava po većem broju ulovljenih mužjaka i drugim problemima prikazanim u istraživanju iz 2010 (Nakládal and Sova 2010.).

KLJUČNE RIJEČI: učinkovitost, feromonski raspršivači, ECOLURE, *Ips typographus*