EFFICACY OF BIOAPIGYN® OINTMENT FOR PELVIC MUSCLE TONUS IN THE TREATMENT OF STRESS URINARY INCONTINENCE AND VULVO-VAGNAL DISORDERS

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ABSTRACT

***Objectives**: The purpose of this work was development, formulation and testing of new herbal ointment for the treatment of stress urinary incontinence and its related symptoms and comparison of its efficiency with the results of pelvic muscle training. The influence of the treatment or training onto vulvo-vaginal disorders was also determined.*

***Materials and methods**: 132 women in the age range from 32 to 77 years were randomly selected into experimental and control group. The experimental group was treated two weeks with Bioapigyn® ointment for pelvic muscle tonus (2 g/day) which consisted of the following ingredients: honey; glycerol, Cera flava, oil macerates of the plants Capsella bursa-pastoris, Urtica diodica, Quercus robur, Salvia officinalis, Achillea millefolium, Alchemilla vulgaris, Calendula officinalis, Matricaria chamomilla, Plantago major; essential oils of the plants Melaleuca alternifolia, Thymus vulgaris ct. tymol, Origanum vulgare. Control group was subjected to pelvic muscle training during 24 weeks (three times a week). The degree of incontinence and its impact on the quality of life prior and after the treatment or training was assessed by the International Consultation on Incontinence Questionnaire - Urinary Incontinence Short Form (ICIQ-UI SF), where maximum score of 21 represents permanent incontinence and 0 no leakage of urine. The volume of residual urine, perineometry and vaginal pH were also determined before and after the treatment or training. For statistical evaluation STATISTICA 11.0 package was used.*

***Results:** The variables with the highest, statistically significant influence onto degree of incontinence and its impact on the quality of life were body mass index and menopause. Significant decrease following the treatment with Bioapigyn® ointment was found for ICIQ-UI SF score (from 12.43±4.83 to 8.61±4.92, p<0.00001), residual urine volume (from 8.73±11.18 to 2.78±5.93 mL, p=0.0002) and vaginal pH (from 6.30±0.63 to 5.59±0.50, p= 0.0000). The increase in the muscle strength was also confirmed. In the end of the treatment all the symptoms of vulvo-vaginal disorders disappeared completely in all 66 participants. Control group also showed some improvement in the values of ICIQ-UI SF score, residual urine and perineometry results. However, those changes were not statistically significant. There was no improvement*

concerning the vaginal pH and the symptoms of vulvo-vaginal disorders.
Conclusion: *Preliminary study confirmed the efficiency* ***of*** *Bioapigyn® ointment for pelvic muscle tonus in the treatment of stress urinary incontinence as well as the symptoms of vulvo-vaginal disorders due to its components with smooth muscle contraction activity, uterotonic activity as well as pH adjusting, coating, moisturizing and soothing effect. Significantly better results for most of the variables were found in the experimental compared to the control group.*

Key words: *stress urinary incontinence, vulvo-vaginal disorders, menopause, vaginal pH, honeybees products, plant extracts, essential oils*

1. INTRODUCTION

Stress urinary incontinence (SUI) is defined as the complaint of involuntary leakage of urine during effort or exertion, or on sneezing or coughing (Luber, 2004). It occurs at least once a week in one third of adult women (Nygaard and Heit, 2004). The prevalence of SUI varies depending on the region of the world and usually ranges between 4% and 35%. Luber, 2004 reported age, obesity, and smoking as the most significant risk factors for development of SUI. The prevalence of SUI among Chinese women was 18.9% (Zhu et al., 2009) and identified risk factors were age, vaginal delivery, multiparty, alcohol consumption, central obesity, constipation, chronic pelvic pain, history of respiratory disease, gynecological events, pelvic surgery, and perimenopause and postmenopause status. 12.4% of French females reported SUI with the pregnancy, particularly previous vaginal delivery and hysterectomy as the significant risk factors (Peyrat et al., 2002). The prevalence of SUI reported by Minassian et al., 2008 among American women was 23.7% with age, ethnic background, weight, parity and hysterectomy as the significant risk factors. The study conducted by Danforth et al., 2006 showed even higher percentage of SUI (up to 43%) among American women. The identified risk factors were age, race/ethnicity, body mass index, parity, smoking, type 2 diabetes mellitus, and hysterectomy. Based on a worldwide study conducted by McPhil, 2004 the highest percentages of women with SUI was found in UK (41%) and Canada (42%) and the lowest in Spain (23%) while the mean value for all tested countries was 32%. Higher prevalence of SUI (Brown, et al., 1999) was obtained among postmenopausal women (56%).

In addition to SUI majority of menopausal women also suffer from vulvo-vaginal disorders triggered by low level of circulating estrogen which causes the changes in the vaginal epithelial cells and consequently leading to the increase in vaginal pH that all together contribute to vaginal dryness, dyspareunia as well as the symptoms like burning, itching, vaginal discharge and unpleasant odor (Pandit and Ouslander, 1997; Calleja-Agius and Brincat, 2015).

The most common treatment approaches for SUI are pelvic floor muscle training, bladder training, vaginal devices, and urethral inserts. Better results were obtained by surgical treatment. However, those methods were associated with more risk compared to the conventional treatment (Nygaard and Heit, 2004).

The purpose of this work was development, formulation and testing of new herbal ointment for the treatment of stress urinary incontinence and its related symptoms as possible alternative to the conventional or surgical methods as well as the comparison of its efficiency

with the results of pelvic floor muscle training. The efficacy of the ointment in the alleviations of the symptoms of vulvo-vaginal disorders was also assessed.

2. MATERIALS AND METHODS

2.1. Study design

The study was designed as the open labeled, randomized, controlled clinical trial. The study protocol was approved by the Ethics Committee of Findri Gustek Health Center. All the participants signed informed consent and completed the questioners.

2.2. Patients

The inclusion criteria for recruitment to the study were history of vaginal delivery, stress urinary incontinence, normal cell cytology, negative urine culture, negative cervical swabs to aerobic bacteria, yeasts, *Ureaplasma urealyticum, Chlamydia trachomatis, Mycoplasma,* and hrHPV DNA, no injuries and bleeding in the vaginal canal, introitus and vestibule. The exclusion criteria were severe prolaps and damage of the recto-vaginal fascia, patients with urge incontinence, patients with severe neurological conditions associated with incontinence, neurogenic bladder, insulin-dependent diabetes mellitus, urinary tract infection, hematuria, age ≤18, pregnancy, less than 24 weeks after vaginal delivery.

2.3. Preparation of the macerate

All plant material was purchased from the certified supplier (Suban, Strmec Samoborski, Croatia). The quality of herbal material used for the macerate production was in accordance with Ph. Eur. criteria requirements. The macerate was prepared from the dried plants and sunflower oil with solid/liquid ratio = 1:5. For the production of the macerate the following plants were used: 20% of areal part of shepherd 's purse (*Capsella bursa-pastoris* L.), 20% of nettle leaves (*Urtica diodica* L.), 10% of oak bark (*Quercus robur* L.), 10% of sage leaves (*Salvia officinalis* L.), 10% of areal parts of yarrow (*Achillea millefolium* L.), 10% of lady's mantle leaves and steam (*Alchemilla vulgaris* L.), 5% of marigold flowers (*Calendula officinalis* L.), 5% of chamomile flowers (*Matricaria chamomilla* L.), 5% of plantain leaves (*Plantago major* L.), 5% of olive leaves (*Olea europaea* L.). Above mentioned proportion of each dried plant was added into stainless steel vessel and mixed thoroughly in order to achieve homogeneity of the sample. After addition of sunflower oil the sample was mixed once again, sealed and extracted by ultrasound for 24 h. Solid/liquid separation was done by filtration.

2.4. Preparation of the ointment

70% of the macerate, 10% of honey, 10% of *Cera flava* (Kemig, Zagreb, Croatia) and 9% of glycerin were heated until 60^0C, mixed slowly for 10 minutes and allowed to cool to 30^0C. Pharmaceutical grade essential oils (Pranarom International, Belgium) of *Melaleuca alternifolia* (0.5%), *Thymus vulgaris* ct. Thymol (0.3%), and *Origanum vulgare* L (0.2%) were added into the

mixture, mixed thoroughly and packed into Alu-tubes of 50 mL volume.

The final product contains: 10% of honey, 10% of *Cera flava,* 9% of glycerin, 14% of the oil macerate of *Capsella bursa-pastoris* L.*,* 14% of the oil macerate of *Urtica diodica* L.*,* 7% of the oil macerate of *Quercus robur* L., 7% of the oil macerate of *Salvia officinalis* L.*,* 7% of the oil macerate of *Achilea millefolium* L.*,* 7% of the oil macerate of *Alchemilla vulgaris* L., 3.5% the oil macerate of *Calendula officinalis* L.*,* 3.5% the oil macerate of *Matricaria chamomilla* L.*,* 3.5% of the oil macerate of *Plantago major* L.*,* 3.5% of the oil macerate of *Olea europaea* L.; essential oils*:* 0.5% of *Melaleuca alternifolia*, 0.3% of *Timus vulgaris* ct. Thymol, 0.2% of *Origanum vulgare.*

2.5. Treatment protocol

Following the exclusion criteria total of 132 patients in the age range from 32 to 77 years with SUI were randomly selected into two groups each of 66 participants. The experimental group was treated with herbal ointment (2 g/day) for two weeks. The ointment was inserted deep into vagina before bedtime using appropriate applicator. The patients were advised to make daily notes about all the changes occurred during the course of the therapy.

The control group of the participants was subjected to pelvic floor muscle training at home, without a health professional's supervision after detailed instruction provided by gynecologist. In brief the participants combined 8-10 second contractions with 5 seconds of rest for 15 minutes, twice daily, 3 times a week for a total duration of 24 weeks.

The interview of each patient was done before and in the end of the treatment. The degree of incontinence and its impact on the quality of life prior and after the treatment was assessed by the International Consultation on Incontinence Questionnaire - Urinary Incontinence Short Form (ICIQ-UI SF), where maximum score of 21 represents permanent incontinence and 0 no leakage of urine. The results of the ICIQ-UI SF may be divided into the following four severity categories: slight (1-5), moderate (6-12), severe (13-18) and very severe (19-21). The residual urine volume was measured before the treatment and in the end of the treatment immediately after the patient returned from the toilet. Measurements were performed with a DC-8 ultrasound unit (Mindray, China). For the measurement of the muscle strength of the pelvic diaphragm, an Apimedis perineometer (EXTT-101, Korea) was used to determine maximal and average pressure (mm Hg) and the mean duration of the contractions (seconds). Vaginal pH was determined before and following the treatment using Woman Vaginal health BV rapid test kit (Assure Tech, Zhejiang, China).

2.6. Statistical Analysis

For statistical evaluation Statistica 11.0 software package was employed. The number of the participant was calculated by Power analysis. With a moderate size effect (0.35), the power strength of 80% and the Type I error rate of 0.05 required sample size was 66 participants per group. The description of the treated population was done by basic statistics and frequency tables. Statistical significance was set to $p<0.05$ in all the tests performed. The differences in the mean value of each parameter prior and after the treatment were assessed by t test and

between control and experimental group by $\chi 2$ test. Pearson correlation coefficients were used for assessing possible correlation between vaginal pH and the age of the participants. The influence of the predictor variables on the degree of the incontinence was tested by Multiple regression method (Orescanin et al., 2015a).

3. RESULTS AND DISCUSSION

3.1. Description of the Population

The participants of the control group (Table 1) ranged from 32 to 74 years (51.79±9.84) with body mass index in the range of 19.84 to 34.25 kg/m^2 (25.7±3.1 kg/m^2) while the number of childbirth ranged from 1 to 5 (2.6±1.4). Among the participants 44 of 66 of them were menopausal or postmenopausal women. All of them suffered from vulvo-vaginal disorders accompanied with at least two symptoms like vaginal dryness, burning, itching, vaginal discharge, painful intercourse and unpleasant odor.

The age range of the experimental group was from 34 to 77 years (59.65±9.37), body mass index ranged from 19.53 to 36.05 (28.59±4.18) while number of childbirth varied from 1 to 4 (2.3±1.2). 52 of 66 participants were menopausal or postmenopausal females that suffered from the same vulvo-vaginal disorders and symptoms as described for control group.

With the onset of menopause the lack of circulating estrogen causes significant changes of the vaginal epithelium including atrophy, dryness, irritation and itching, incontinence as well as increase in vaginal pH that could if not treated lead to both urinary tract infections as well as lower genital tract infections (Pandit and Ouslander, 1997; Calleja-Agius and Brincat, 2015).

Table 1. The basic statistical parameters for age, body mass index (BMI) and number of childbirths of the control and experimental group

Variable	Statistical parameter					
	Control group			Experimental group		
	$\bar{X}$±SD	Min.	Max.	$\bar{X}$±SD	Min.	Max.
Age (years)	51.79±9.84	32.00	74.00	59.65±9.37	34.00	77.00
BMI (kg/m^2)	25.7±3.1	19.84	34.25	28.59±4.18	19.53	36.05
No. of childbirth	2.6±1.4	1	5	2.3±1.2	1	4

The results of multiple regression analysis (Table 2) testing for the correlation between International Consultation on Incontinence Questionnaire - Urinary Incontinence Short Form (ICIQ-UI SF) score as dependent variable and selected predictor variables of the control population showed good, statistically significant correlation (R= 0.62; p<0.0273) with body mass index and menopause as the only statistically significant variables that contributed the most to the overall correlation. Although, somewhat higher values of ICIQ-UI SF values were found in the group older than 50 years (8.61±4.34) compared to the younger group (8.30±4.02) as well as those with three or more childbirth (8.52±4.28) compared to the participant with one and

two childbirth (8.31±4.27) those differences were not statistically significant (p=0.2031 and p=0.3744 for age and number of childbirth, respectively).

The results of multiple regression analysis performed on the experimental group (Table 2) between ICIQ-UI SF score as dependent variable and selected predictor variables showed even better, statistically significant overall correlation compared to the control group (R= 0.74; p<0.0198). Similar to the control group the only statistically significant predictors were BMI (p= 0.0278) and menopause (p=0.0326). The value of ICIQ-UI SF score increased with age of the participant as well as the number of childbirth but these variables showed no statistically significant contribution to the correlation.

Our results obtained for both control and experimental group were in agreement with previous research that identified obesity, perimenopause and postmenopause status, age and vaginal delivery as most common risk factors for the development of SUI (Brown, et al., 1999; Peyrat et al., 2002; Luber, 2004; Danforth et al., 2006; Minassian et al., 2008; Zhu et al., 2009).

Table 2. The results of multiple regression analysis testing for the correlation between International Consultation on Incontinence Questionnaire - Urinary Incontinence Short Form (ICIQ-UI SF) score as dependent variable and selected predictor variables for the control population. .*-statistically significant at p<0.05

Predictor variable	Statistical parameter			
	Control group		Experimental group	
	β	p	β	p
No. of childbirth	0.18	0.3933	0.22	0.1928
Age	0.16	0.4244	0.18	0.4140
BMI	0.46	0.0384*	0.51	0.0278*
Menopause	0.41	0.0401*	0.49	0.0326*
	R= 0.62; p<0.0273*		R= 0.74; p<0.0198*	

3.2. The outcome of the treatment

Frequencies and percentages for the severity of the symptoms of stress urinary incontinence based on the results of ICIQ-UI-SF score before and following the treatment determined in the control group subjected to pelvic floor muscle training were presented in Table 3. Following the training the patients with slight symptoms increased from 15.15% to 18.18% while those with severe symptoms decreased from 30.30 to 24.24%.

Table 4 presents the basic statistical parameters for International Consultation on Incontinence Questionnaire - Urinary Incontinence Short Form (ICIQ-UI SF), perineometry with maximal pressure, average pressure and duration of the pressure, residual urine volume and vaginal pH determined in the control group prior and after the pelvic floor muscle training. After completion of six months of the training the mean value of ICIQ-UI SF score slightly decreased from 8.34±4.21 to 7.92±4.71 and so was the residual urine volume (from 5.12±3.73 to 4.52±3.64). However, according to t-test this decrease was no statistically significant for neither of these two variables.

Table 3. Frequency table for the severity of the symptoms of stress urinary incontinence based on the results of ICIQ-UI-SF score before and following the pelvic floor muscle training

Symptoms (score)	Before the training		Following the training	
	N	%	N	%
Slight (1-5)	10	15.15	12	18.18
Moderate (6-12)	36	54.55	38	57.58
Severe (13-18)	20	30.30	16	24.24

Perineometry results (Table 4) showed some increase in the strength of pelvic musculature which was reflected in the increase of maximal pressure (from 15.19±9.01 to 17.54±8.23 mm Hg, average pressure (from 9.37±7.71 to 11.84±7.63 mm Hg) as well as the duration of the pressure (from 15.1±10.6 to 16.2±13.2 s). However, based on the results of t-test none of those three variables showed statistically significant increase.

Vaginal pH (Table 4) showed almost no changes following the pelvic floor muscle training (6.41±0.71 and 6.38±0.69 before and after the training) since there was no medical treatment of any kind during those six months. Expectedly, there was also no improvement in the vulvo-vaginal complaints accompanied with the symptoms like vaginal dryness, burning, itching, vaginal discharge, painful intercourse and unpleasant odor that could be directly linked with high pH values and low serum estradiol level.

pH value was significantly higher compared to the values found in healthy, reproductive age females which usually ranges from 3.6 and 4.5 (Boskey et al., 1999). Higher values could be the sign of either bacterial vaginosis (BV) (usually in reproductive age women) or clear sign of menopause. Since 66.7% of the participants were either menopausal or postmenopausal women and BV was excluded high pH value could be directly linked with menopause. Indeed premenopausal women from our study had mean pH value 5.01±0.51 while menopausal and postmenopausal had vaginal pH of 6.61±0.53. Besides, Pearson correlation coefficient between vaginal pH and age showed very good, statistically significant correlation (r=0.72; p=0.0021). Previous studies suggested that vaginal pH could be used as an indicator of menopause since its correlated well with the level of serum follicle stimulating hormone (FSH) - a standard diagnostic tool for menopause confirmation (Milsom et al., 1993; Roy et al., 2004; Panda et al., 2014). Panda et al., 2014 reported the mean vaginal pH of 5.3 ± 0.7 for 173 menopausal women ranging from 31 to 60 years with 84.9% sensitivity. Pandit i Ouslander, 1997 reported that vaginal pH >5 has the sensitivity of 64-67% for the confirmation of menopause. Cailloutte et al. 1997 assessed vaginal pH and serum FSH level among 172 postmenopausal women. They reported that the sensitivity of vaginal pH in predicting estradiol status was 88%. Vaginal pH of 4.5 and lower is consistent with a premenopausal serum estradiol level and the absence of bacterial vaginosis. Increase of vaginal pH between 5.0 and 6.5 could be connected either with bacterial vaginosis or decreased serum estradiol level while in the absence of BV, vaginal pH of 6.0 to 7.5 could be strongly linked with menopause. In the absence of BV, vaginal pH >4.5 indicated menopause with a sensitivity of 74% (Roy et al. 2004). They reported weighted average vaginal pH of 6.0 for menopausal women who do not receive estrogen therapy. Another study conducted on 103 menopausal women obtained vaginal pH of 5.33±0.53 with 97% sensitivity for menopause confirmation (Vahidroodsari et al., 2010). Tuntiviriyapun et al., 2015

found significant correlation between vaginal pH and vaginal atrophy symptoms by conducted the study on 132 naturally postmenopausal women.

Table 4. The basic statistical parameters for International Consultation on Incontinence Questionnaire - Urinary Incontinence Short Form (ICIQ-UI SF), perineometry with maximal, average and duration of the pressure, residual urine volume and vaginal pH determined in the control group subjected to pelvic floor muscle training prior and after the training.*-statistically significant at p<0.05

Variable	Before the training			Following the training			t-test	
	$\bar{X}$±SD	Min.	Max.	$\bar{X}$±SD	Min.	Max.	t	p
ICIQ-UI-SF	8.34±4.21	4.00	18.00	7.92±4.71	4.00	17.00	0.5	0.5900
Max. pressure (mm Hg)	15.19±9.01	0.00	23.00	17.54±8.23	0.00	26.90	1.6	0.1201
Average pressure (mm Hg)	9.37±7.71	0.00	19.00	11.84±7.63	0.00	22.74	1.9	0.0666
Duration of pressure (s)	15.1±10.6	0.00	26.00	16.2±13.2	0.00	29.40	0.5	0.5985
Residual urine (mL)	5.12±3.73	0.00	10.07	4.52±3.64	0.00	8.43	0.9	0.3514
Vaginal pH	6.41±0.71	5.00	7.50	6.38±0.69	5.00	7.50	0.2	0.8059

3.2 Experimental group

Following two weeks of the treatment with Bioapigyn® ointment for pelvic muscle tonus resulted in decrease of the severity of the symptoms of SUI. After the treatment 4.56% of the patients were completely dry while very severe symptoms decreased from 10.61% to 0 and severe symptoms from 42.42% to 25.75% (Table 5).

Significant decrease following the treatment with Bioapigyn® ointment for pelvic muscle tonus (Table 6) was found for ICIQ-UI SF score (from 12.43±4.83 to 8.61±4.92, p<0.00001), residual urine volume (from 8.73±11.18 to 2.78±5.93 mL, p=0.0002) and vaginal pH (from 6.30±0.63 to 5.59±0.50, p= 0.0000). Similar to the control group pH value was lower in premenopausal (5.25±0.31) compared with menopausal and postmenopausal women (6.47±0.34). Excellent correlation was obtained between vaginal pH and age of the participant (r=0.91; p=0.0000). The pelvic muscle strength also showed improvement following the treatment with increase of maximal pressure from 9.87±11.00 to 12.37±11.26 mm Hg, average pressure from 6.21±8.41 to 7.94±7.84 mm Hg as well as in the duration of the contraction from 5.65±7.59 to 7.44±9.51 s. However, this increase was not statistically significant which could be partially explained with high deviation among the participants.

All the participants reported improvement which was reflected in lower number of urination during the day and night, the possibility of complete emptying of the bladder half an hour after the application of the ointment, ability to retain the urine much longer, lower volume and frequency of the escaped urine during coughing, sneezing and physical activity.

Table 5. Frequency table for the severity of the symptoms of stress urinary incontinence based on the results of ICIQ-UI-SF score before and following the treatment determined in the experimental group

Symptoms (score)	Before the treatment		Following the treatment	
	N	%	N	%
None (0)	0	0	3	4.56
Slight (1-5)	7	10.61	20	30.30
Moderate (6-12)	24	36.36	26	39.39
Severe (13-18)	28	42.42	17	25.75
Very severe (19-21)	7	10.61	0	0

Table 6. The basic statistical parameter for International Consultation on Incontinence Questionnaire - Urinary Incontinence Short Form (ICIQ-UI SF), perineometry with maximal, average and duration of the pressure, residual urine volume and vaginal pH in determined in the experimental group prior and after the treatment.*-statistically significant at p<0.05

Variable	Before the treatment			Following the treatment			t-test	
	$\bar{X}$±SD	Min.	Max.	$\bar{X}$±SD	Min.	Max.	t	p
ICIQ-UI-SF	12.43±4.83	5.00	21.00	8.61±4.92	0.00	18.00	4.5	<0.00001*
Max. pressure (mm Hg)	9.87±11.00	0.00	47.00	12.37±11.26	0.00	34.20	1.2	0.2013
Average pressure (mm Hg)	6.21±8.41	0.00	39.00	7.94±7.84	0.00	28.70	0.7	0.3864
Duration of pressure (s)	5.65±7.59	0.00	29.00	7.44±9.51	0.00	30.00	1.1	0.2342
Residual urine (mL)	8.73±11.18	0.00	33.05	2.78±5.93	0.00	17.4	4.0	0.0002*
Vaginal pH	6.30±0.63	5.00	7.50	5.59±0.50	4.50	6.50	4.1	0.0001*

Those results were in concordance with the previous study using very similar formulation of the ointment (Oreščanin and Findri Guštek, 2016). After two weeks of the application of 2.0 g of ointment once a day the mean value of ICIQ-UI SF score decreased from 10.3±4.2 to 7.2±1.8 (30.1%).

Significant improvement following the therapy in the present study could be explained by the composition of the ointment containing the plant macerate of *Capsella bursa-pastoris* and *Urtica diodica* that induced smooth muscles contraction (Orescanin and Findri Gustek, 2016; Al-Snafi, 2015; Grosso et al., 2011; Broncano et al., 1987), and consequently, enhanced uterine muscle strength significantly. Besides, **t**he plants with confirmed uterotonic activity (*Matricaria chamomilla*, *Calendula officinalis*, *Plantago major*, *Capsella bursa pastoris*) and astringent properties (*Quercus robur, Achillea millefolium*, *Salvia officinalis*, *Olea europaea*, *Plantago major*) resulted in the tightening and firming of the smooth muscles of the pelvic floor and consequently, reduced the symptoms of incontinence significantly especially in perimenopausal and menopausal women (Shipochliev, 1981; EMA, 2009; Gruber et al., 2011; Orescanin and Findri Gustek, 2016).

In addition to the decrease of SUI symptoms, all the symptoms of vulvo-vaginal

disorders disappeared after two weeks of the application of the ointment with pH 4.46 which could be connected with significant decrease of vaginal pH and re-establishment of healthy balance of the vaginal flora. This could be mostly connected with the presence of honey in the product. Honey with its low pH value, prebiotic and probiotic activity and osmotic effect of sugars encouraged the development of normal vaginal flora and established normal pH of the vagina for menopausal/postmenopausal women (Orescanin and Findri Gustek, 2015 a,b,c).

Furthermore, the plants macerate in the combination with essential oils maintained the healthy balance of very complex vaginal flora by preventing uncontrolled growth of the pathogens and also served as natural preservatives in the ointment as well as odor correctors (Orescanin and Findri Gustek, 2015a,b,c).

Cera flava prevented irritations of the genital tract membranes by forming protective layer while glycerol in the combination with honey provided excellent moisturizing effect which reduced vaginal dryness and pain (Orescanin and Findri Gustek, 2015 a,b,c).

The purpose of this study was also to compare the efficiency of Bioapigyn® ointment for pelvic muscle tonus in relieving the symptoms of SUI compared to pelvic floor muscle training. Since direct comparison of the mean values of selected variables wouldn't be appropriate approach due to differences in the initial values in two groups the percentage of the changes (decrease or increase) of the mean value of each variable following the treatment was calculated for both groups and the differences between the percentages were tested by χ^2 test (Table 7). It was clearly visible that the experimental group showed better results for all tested variables. The differences between two groups were statistically significant for ICIQ-UI-SF score, duration of the pressure, residual urine volume and vaginal pH.

Table 7. Percentage of the changes (decrease or increase) of the mean value of each variable following the treatment with Bioapigyn® ointment for pelvic muscle tonus (experimental group) or pelvic muscle training (control group) and statistical significance between experimental and control group

Variable	Changes of the variable after the treatment or training (%)		p
	Experimental group	Control group	
ICIQ-UI-SF	30.7	5.0	0.0001*
Max. pressure (mm Hg)	25.3	15.5	0.1624
Average pressure (mm Hg)	27.9	26.4	0.8464
Duration of pressure (s)	31.7	7.3	0.0004*
Residual urine (mL)	68.2	11.7	0.0000*
Vaginal pH	11.3	0.5	0.0085*

4. CONCLUSIONS

The study was conducted on 132 mostly menopausal or postmenopausal age women that suffered from stress urinary incontinence as well as vulvo-vaginal disorders accompanied with the symptoms like vaginal dryness, burning, itching, vaginal discharge, painful intercourse and unpleasant odor. The experimental group was treated with new Bioapigyn® vaginal ointment for pelvic muscle tonus while the control group was subjected to pelvic floor muscle training. The results of each approach were quantified using ICIQ-UI-SF score, perineometry, residual urine volume and vaginal pH as well as the presents/absence of the symptoms of vulvo-vaginal disorders before and after the treatment/training. In the end of two weeks of the treatment the mean value of ICIQ-UI score for the experimental group decreased 30.7% times, perineometry parameters increased between 25.3 and 31.7%, residual urine decreased for 68.2% and vaginal pH for 11.3%. All the symptoms of vulvo-vaginal disorders disappeared completely in all participants. The participants subjected to pelvic muscle training showed no changes in vaginal pH. There was no improvement concerning the symptoms of vulvo-vaginal complaints. ICIQ-UI score decreased 5%, residual urine volume for 11.7% while perineometry parameters increased between 7.3 and 26.4%. The Bioapigyn® ointment treated group showed significantly better results in ICIQ-UI-SF score, muscle strength expressed as the duration of the pressure, residual urine volume and vaginal pH compared to the control group. Besides, the ointment showed excellent results in the treatment of vulvo-vaginal disorders in menopausal and postmenopausal women.

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