A trial to prove the efficacy of acupuncture as a therapeutic support in pharmacological prophylaxis for migraine and tension-type headache. Pilot study

*Sebastiano Olivier, Annibale Antonioni, Beatrice Mezzetta, Jay Guido Capone, Maura Pugliatti, Enrico Granieri**

Section of Neurology, Psychiatry and Psychology, Department of Neuroscience and Rehabilitation, University of Ferrara, Ferrara, Italy. * E-mail: enrico.granieri@unife.it

Abstract: *Importance:* the effectiveness of acupunture as a therapeutic support in pharmacological prophilaxis for migraine and tension-type headache. *Objective:* to define the role of acupuncture in the treatment and prophylaxis of various types of migraine by measuring the effects it produces in preventing the disease, reducing symptoms and their prolongation over time. *Design, setting and partecipants:* this work, which may be considered a pilot study, provides the clinical results obtained in a period of four months from September 2018 to September 2019. The 42 participants, recruited from patients of the Headache Center of the Neurology Unit of Sant'Anna University Hospital of Ferrara, were randomly divided into two groups. Group A received pharmacological prophylaxis (Control group), while Group B undertook pharmacological prophylaxis and acupuncture (Experimental group). *Interventions:* The control group took medication for four months, while the experimental group received a twice-weekly treatment of eight-twenty minutes acupuncture sessions every two weeks. *Main outcome and measures:* all participants were examined before the start of prophylaxis (T0), after two months (T1) and after four months (T2). They were asked to evaluate the daily progress of their psycho-physical conditions in a diary, from which it emerged that the practice of acupuncture reduced not only the frequency and intensity of daily attacks, but also the use of analgesics, revealing a prolongation of the positive effects for four months. *Results:* Group B showed a significant reduction in the Henry Ford Headache Disability Inventory - beta (β-HDI) score in both the second and fourth month compared to Group A, with a similar pattern emerging for the Migraine Disability Assessment Score (MIDAS). Likewise, an improvement in physical conditions and a decrease in pain were recorded as far as the Short Form 36 Health Survey (SF-36) scale was concerned. *Conclusions and relevance:* this study shows that acupuncture is a valid supportive therapy for pharmacological prophylaxis of migraine and tension-type headache.

Key words: Headache, Acupuncture, Migraine, Unconventional therapies, Headache prophylaxis

Introduction

Headache is the most common neurological symptom. Despite its incidence, or perhaps precisely because of it, headaches have always been underestimated or misunderstood. The risk is that headaches may not be correctly diagnosed and, consequently, inadequately treated, leading to varying degrees of repercussions on many aspects of life. In its various forms, headache is disabling and often generates fears of serious underlying diseases (1). Headache has a world prevalence of 47% in the adult population with important negative effects on quality of life, productivity and the economy. The most common forms are tension-type headache (38%), migraine (10%) and chronic headaches (3%). Headache is the second leading cause of "years lived with disabili-

ties" worldwide and migraine alone is the third leading cause in the population aged between 15 and 49. From a global perspective, the indirect costs it entails, due to the loss of working hours, represent a heavy burden for the community (2, 3). A recent review (2010) revealed that the prevalence of headaches has greatly increased in Europe in recent decades. For tension-type headache there is a prevalence of 60%, 15% for migraine, 4% for chronic headaches, a possible 1-2% for drug overuse headaches and 0.2-0.3% for cluster headaches. Headache is prevalent in both sexes and more frequent in adulthood; however, higher prevalences occur in women at an age ranging from 20 to 50, suggesting a possible role for hormonal factors (4). A recent study carried out in Italy reports how the reduction in productivity, drug intake, diagnostic procedures, non-pharmacological treatments and other factors associated with headache involve an annual cost pro capite estimated at over € 10,000 (5, 6). Tension-type headache compared to migraine is also more related to a series of psychosocial variables. The results of various studies suggest, instead, that migraine is mainly a constitutional disorder (7, 8).

Thanks to the advances in research and training, acupuncture can now be considered a valid option for those suffering from headache. Recent evidence on the action of acupuncture has shown that the skin areas corresponding to certain acupoints are particularly rich in sensitive endings. The dynamics of tissue homeostasis seems to lead to painful sensitisation of these areas and that the insertion of needles has an action on the Ca^{2+} ions channels involved in the release of β-endorphins (9). Although the localisation of acupoints and meridians is the result of empirical observations, scientific studies indicate that there are underlying neurophysiological mechanisms (10). Acupuncture is considered 'complementary' (or 'alternative') medicine, ie., involving the controlled and scientifically approved use of health methods which are not usually considered part of conventional therapy. It may therefore represent a minimally invasive and low-risk method used as 'add-on' to common drugs (11).

A 2012 meta-analysis measured the effectiveness of acupuncture with respect to sham acupuncture and no treatment in chronic pain for four pathological conditions, including migraine. In this study there is evidence on the effectiveness in easing the intensity of pain. Acupuncture is defined as a reasonable prophylactic option for chronic migraine (12). Furthermore, two more recent Cochrane meta-analyzes provide reliable results on the efficacy of acupuncture as a prophylaxis for migraine and tension-type headache. Prophylactic treatment with acupuncture was evaluated with respect to symptomatic drug treatment alone, drug prophylaxis (a physical/physiotherapeutic prophylaxis in tension-type headache) and sham acupuncture. The authors highlighted that acupuncture is effective in reducing the frequency of migraine and tension attacks compared to symptomatic treatment with drugs alone. Studies have also shown that acupuncture has a prophylactic efficacy and can therefore be considered a viable therapeutic option in headache prophylaxis, but with a lower level of confidence (13). In 2015, a randomized controlled Italian study analyzed data from 131 migraine sufferers recruited from the Headache Centers in Bologna and Parma. The comparison between drug prophylaxis and acupuncture prophylaxis shows that acupuncture is effective in reducing the frequency of attacks and the negative impact of migraines on quality of life (14). The following systematic reviews and meta-analyzes confirm that acupuncture is associated with good clinical outcomes compared to standard care, especially for short (two months) follow-up. Current evidence supports acupuncture as a potential treatment for patients with migraine, chronic migraine and tension-type headache (15).

Outcomes of the study

The purpose of our prospective study was to evaluate the role of acupuncture therapy for prophylactic purposes in patients suffering from migraine (with and without aura, chronic) with indications to pharmacological prophylaxis, taking into consideration the possible coexistence of tension type headache (episodic, frequent episodic). The primary outcomes of the study were the evaluation of the intervention in terms of:

- Monthly frequency of attacks (days / month)
- Average monthly intensity of attacks
- Monthly intake of analgesics
- Monthly intake of triptans
- Average monthly disability

The secondary outcomes were the impact of the headache on:
- Trait anxiety disorder
- Depressive disorder
- General health status

Materials and Methods

Study design and intervention methodology

Patients who met the inclusion criteria were assigned to two groups: drug prophylaxis (Group A) and drug prophylaxis + acupuncture prophylaxis (Group B). Most of the subjects included in both group A and group B underwent pharmacoprophylaxis for migraine with Topiramate and Lamotrigine, a few were given Beta-blockers (Propranolol), particularly young subjects, and only two subjects were treated with periodic injections of botulinum toxin. In Group A, the patients underwent a drug prophylaxis treatment lasting at least four months. The Group B patients underwent a cycle of eight acupuncture sessions lasting 20 minutes each every two weeks. Once in place, the needles were not manipulated until they were removed. The treatment was carried out by a single acupuncturist on the basis of a previously developed diagnostic-therapeutic protocol. The patients of this group, all with indications to undertake a drug prophylaxis of at least four months, were given the choice of whether to undertake it or not. All subjects were evaluated by a single operator before starting the prophylaxis (baseline-T0), after two months (T1) and after four months (T2). On all three occasions, their headache diaries were evaluated and self-assessment questionnaires were administered to investigate multidimensional aspects related to headache and its repercussions on daily life. At the first meeting all patients were subjected to an ad hoc questionnaire consisting of three parts:
- The first part concerned personal and anamnestic data
- The second part was devoted to headache with references to clinical characteristics (age of onset, temporal course, frequency, related disability, intensity, quality and location of pain, accompanying symptoms, triggers, drug history)
- The third part (reserved only for patients of Group B) represented the section dedicated to acupuncture, with specific data for the therapeutic choice by the acupuncturist.

Patient meetings took place in Ferrara, in the medical office of Dr. Beatrice Mezzetta, a physician-surgeon specialized in acupuncture. When this was not possible, patients were contacted by telephone or online to describe the study and verify their willingness to participate; later, the same means could be used to obtain information about the progress of symptoms after the start of therapy and/or to fill out some of the questionnaires. The acupuncture sessions, with a preliminary visit, were carried out by Dr. Mezzetta Beatrice in her office.

This study was conducted in accordance with World Medical Association Declaration of Helsinki and received approval from the Ethics Committee of the University of Ferrara.

Patient recruitment

On the recommendation of a neurologist, subjects were recruited among those who attended the Headache Center of the Neurology Operating Unit of the Sant'Anna University Hospital of Ferrara or the neurological clinics of the Local Health Unit of Ferrara - Casa della "Salute Cittadella S. Rocco", over a period of time from September 2018 to September 2019.
As part of a pilot study, and considering the possibility of limited resources, time, and numbers, by alternating assignment of consecutive sets of patients in Group A and Group B a randomization procedure was chosen, in the assumption that there would be homogeneity among the patients belonging to the centers where recruitment was carried out. As for information and acceptance of consent, each patient was interviewed, in person or by telephone, to confirm their candidacy and was informed of the objectives and procedures of the study. Participation in the study was formalized by signing an informed consent form.

Inclusion criteria

- Both males and females
- Age from 18 to 65 years

- A headache diary for the previous three months
- Neurological diagnosis of migraine without aura, migraine with aura or chronic migraine, and / or infrequent episodic tension-type headache, frequent episodic tension-type headache (according to the criteria of the International Classification of Headache Disorders, ICHD-3 2018)
- Indication for pharmacological prophylaxis for at least four months, with a four-day minimum of disabling headache

Exclusion criteria

- State of pregnancy
- Medical conditions identifiable as causes of secondary headaches
- Documented overuse of symptomatic drugs in the three months prior to neurological diagnosis
- Severe psychiatric morbidity

In addition, for Group B:

- HIV, HBV, HCV infection
- Coagulation disorders or therapy with oral anticoagulants.

Outcome measures

- Headache diary: A headache diary is the most effective tool for a correct diagnosis and monitoring of headache. Once a prophylaxis therapy has started, filling in the diary allows us to monitor the effects of the treatment. In addition to the monthly frequency of days and attacks, the following data can be recorded for each headache episode: type of headache, location, quality, intensity and evolution of pain, duration of the attack, disability related to the attack, symptomatic medication intake, any favoring factors (16). For this study, a diary was prepared to provide data on: frequency of attacks (days / month) and within 24 hours: type of attack, intensity (Numeric Rating Scale for pain intensity; 0 no pain -10 worst possible pain), related disability (0 absent; 1-mild; 2-moderate; 3-severe) and symptomatic medication intake (number of doses of analgesics and triptans) (17-19). At the time of enrollment, there were patients who were already keep a headache diary; during the four months of the study they continued to use that diary.
- Self-assessment questionnaires: In this study, five of the most accredited self-evaluation questionnaires in the field of headache research (Henry Ford Headache Disability Inventory-beta, Migraine Disability Assessment (MIDAS), State-Trait Anxiety Inventory: Y Form, Beck Depression Inventory-II (BDI-II) and Short Form-36 Health Survey) were administered. The questionnaires and evaluation scales were chosen to investigate the impairment of quality of life in relation to headache and its possible interactions with anxiety disorder and depressive disorder (20,21). As far as the the timing of assessment was concerned, each questionnaire was administered to patients in the three periods of the study, with the exception of the Migraine Disability Assessment (MIDAS), which was only administered at T0 and T1.
- Henry Ford Headache Disability Inventory - beta (β-HDI): questionnaire of 25 questions related to headache-related disability. The total score ranges from 0 to 100 according to five categories: 0-9: no disability; 10-29: mild disability; 30-49: moderate disability; 50-71: severe disability; 72-100: complete disability (22,23). The original test (in English) was translated into Italian with the help of a specialized English mother-tongue translator.
- Migraine Disability Assessment (MIDAS): the questionnaire consists of five questions whose answers express the number of days with activities compromised by migraine over three months. The total score is the numerical sum of the responses to the five questions and is categorized into four degrees of disability: 0-5: grade 1- none; 6-10: grade 2- mild; 11-20: grade 3-moderate; ≥21: grade 4- severe (24,25). Italian version: 10.1046/j.0333-1024.2001.00277.x
- State-Trait Anxiety Inventory: form Y (STAI-Y): this includes two scales of 20 questions each. The first scale (STAI-1-state) measures the subject's state of anxiety at the time of compilation. The second (STAI-2-trait), instead, evaluates the relatively "stable" aspects of anxiety (26, 27). Validation of Italian version: Pedrabissi, L., & Santinello, M. (1989). Verifica della validità dello STAI forma Y di Spielberger [Verification of the validity of the STAI, Form Y, by Spielberger]. *Giunti Organizzazioni Speciali, 191-192,* 11–14.
- Beck Depression Inventory-II (BDI-II): this allows us to evaluate the patient's degree of depression and, if

repeated, to monitor the effects of a therapeutic intervention; it consists of 21 questions assessing the common depressive symptoms in terms of severity; each answer corresponds to a specific score and the final score ranges from 0 to 63. There are four categories: 0-13: minimal depression; 14-19: mild depression; 20-28: moderate depression; 29-63: severe depression (28). Italian version: http://hdl.handle.net/11577/1782674

- Short Form 36 Health Survey (SF-36): this multidimensional questionnaire, validated in Italian (29), focusses on the health as perceived by the patient; it consists of 36 questions referring to eight health domains. The results for each subscale are expressed with a range of variability from 0 to 100. All questions refer to the month before completion, except for the question about the change in health status (30).

Statistical analyses

Frequencies and percentages were reported for descriptive purposes for categorical variables and means, and standard deviations (SD) for continuous variables. The comparison analyses were carried out by applying a chi-square test for categorical variables and student-t for continuous variables where a normal type distribution was assumed. For the comparisons of the data relating to the variables taken from the headache diary, and from the HDI, BDI-II, MIDAS, STAI-Trait tests and for the different items of the SF-36 scale, non-parametric tests were used, such as a Wilcoxon test for the comparison of two dependent variables (T1 vs T0) and a Friedman test for the comparison of multiple dependent variables (T2 vs T1 vs T0). For these analyses, the medians of the distributions were reported. The statistical analysis was carried out using the Statistical Package for Social Sciences (SPSS) version 19 for Windows and OSX (SPSS Inc., IBM, Somers, New York, USA).

Results

The study was conducted on 42 subjects, including ten men and 32 women with a mean age (SD) of 45.9 (15.2) years and 44.8 (13.0) years, respectively (p=0.54). Subjects were assigned to Group A and Group B by alternating consecutive series. Table 1 shows the main clinical-demographic characteristics of the sample under study, which do not show statistically significant differences between Group A and Group B. The study sample includes a subgroup of patients suffering from mixed headache with migraine and a tension-type headache: eight were assigned to Group A and seven to Group B. Although, according to the most recent headache classification criteria (ICHD-3), these patients should be classified as suffering from chronic migraine, we have highlighted the fact that they also meet the classification criteria for tension-type headache in order to provide insights into the possible role of acupuncture in this type of headache as well.

Table 1: Description of the study sample by mode of intervention (acupuncture vs. observation)

Table 2 shows the differences observed by examining the various items of the headache questionnaire between the two groups at time 0 (baseline), after two and after four months, respectively, for Group A and Group B. The results refer to the comparison between the 2nd month and baseline, between the 4th month and baseline, considering all three points. Comparison of the medians suggests that acupuncture has a particular effect on the number of headache days per month (frequency), with persistence up to the 4th month, the consequent use of analgesic drugs and triptans, the average intensity of attacks as well as on mean headache-related disabilities (Table 2).

Table 2: Characteristics (medians) from the headache diary: comparison between the two groups (Group B vs A) and the different periods studied (T0, T1, T2)

Compared to the medians, Group B in particular showed a reduction in the HDI score at the 2nd month and even more at the 4th month compared to baseline and Group A (Table 3). A similar trend was observed for MIDAS scores between baseline and the 4th month. In the context of the SF-36 scale, the most evident impact compared to Group A was seen for the "limitation due to physical function", and pain. For the domains of "limitations due to emotional problems", and of "emotional well-being", only Group A showed any benefit, while in neither group was there a change from baseli-

Table 1: Description of the study sample by mode of intervention

	Group A Pharmacoprophylaxis	Group B Pharm+ Acupuncture	*P*
N (%)	22 (52.4)	20 (47.6)	
Gender, female sex (%)	18 (81.8)	14 (70.0)	*0.477*[a]
Age at the time of the study, years, average age (SD)	46.3 (11.9)	43.7 (15.0)	*0.540*[b]
Type oh headache*			
Migraine	14 (63.6)	13 (65.0)	*0.927*[c]
Mixed (Migraine + Tension-Type Headache)	8 (36.4)	7 (35.0)	
Duration of headache from onset, years, average (SD)	29.4 (13.7)	27.8 (12.7)	*0.695*[b]
Duration of headache from clinical aggravation, years, average (SD)	19.3 (12.3)	13.4 (11.2)	*0.112*[b]
Familiarity with headache, N (%)	18 (81.8)	18 (90.0)	*0.665*[a]
Maximum level of education			
None/Primary School: N (%)	1 (4.5)	1 (5.0)	*0.365*[a]
Middle School graduation: N (%)	3 (13.6)	4 (20.0)	
High School Diploma: N (%)	8 (36.4)	11 (55.0)	
Graduation/University Degree: N (%)	10 (45.5)	4 (20.0)	
Employment			
Housewife	2 (9.1)	1 (5.0)	*0.784*[a]
Unemplyed	0 (-)	1 (5.0)	
Manual worker	8 (36.4)	5 (25.0)	
Not manual worker	9 (40.9)	8 (40.0)	
Retired	1 (4.5)	1 (5.0)	
Student	2 (9.0)	4 (20.0)	

*includes a subgroup of patients with mixed headache with migraine-tension component

ne for the SF-36 domains of "physical function", "social functioning" and "general health" (Table 3).

The same analysis was performed in a subgroup of patients who additionally presented tension-type headache. Compared to the group of subjects with migraine headache only, the sample had some significantly different characteristics, such as the average age at the time of the study, lower in subjects with mixed headache, and the distribution of the variables relating to employment.

In general, when comparing the means for the headache diary variables, a significant difference emerged only for some items (monthly headache frequency, intensity and relative disability) in Group B and at 4th month compared to baseline, with a trend in line also in the 2nd month, but not statistically significant (Table 4).

Regarding the tension component, Group B only showed a weak reduction in MIDAS score from baseline to 4th month compared to Group A, and even in the SF-36 domains of "physical function" and "pain" (Table 5). For BDI-II, STAI-Trait scores, and the SF-36 domains of "limitations due to emotional problems" and "emotional well-being", a positive change was observed only in Group A. No other changes were observed over time between the two Groups for all other scores.

Discussion and Conclusion

The hypothesis that inspired the planning of this study is whether, and in what way, acupuncture can represent an effective support to standard symptomatic and, in particular, prophylactic therapies for headaches. A common problem in pharmacological prophylaxis for headaches is compliance, often compromised by the patient's expectations and by the possible side

Table 2. Headache main features from the headache diary: comparison between Group A and Group B at the study baseline, 2-month and 4-month follow up, respectively.

	Group A	Group B
Frequency of attacks (median, n. day, months)		
T_0	7.5	11.0
T_1	7.0	7.0
T_2	6.0	7.0
$p\ (T_0\text{-}T_1)^a$	***0.012***	***0.0004***
$p\ (T_0\text{-}T_2)^a$	***0.010***	***0.001***
$p\ (T_0\text{-}T_1\text{-}T_2)^b$	***0.048***	***0.0001***
Use of analgesics (number/months)		
T_0	2.5	3.0
T_1	2.0	1.0
T_2	1.0	0.5
$p\ (T_0\text{-}T_1)^a$	*0.565*	***0.005***
$p\ (T_0\text{-}T_2)^a$	***0.017***	***0.001***
$p\ (T_0\text{-}T_1\text{-}T_2)^b$	***0.023***	***0.001***
Use of triptans (number/months)		
T_0	5.5	1.0
T_1	4.5	1.0
T_2	3.5	0.0
$p\ (T_0\text{-}T_1)^a$	*0.130*	***0.007***
$p\ (T_0\text{-}T_2)^a$	*0.052*	***0.005***
$p\ (T_0\text{-}T_1\text{-}T_2)^b$	*0.236*	***0.001***
Average intensity (median, n. attacks/month)		
T_0	6.0	6.0
T_1	5.9	4.0
T_2	6.0	3.9
$p\ (T_0\text{-}T_1)^a$	*0.347*	***0.0001***
$p\ (T_0\text{-}T_2)^a$	*0.063*	***0.001***
$p\ (T_0\text{-}T_1\text{-}T_2)^b$	*0.614*	***0.00007***
Average disability/months		
T_0	2.0	2.0
T_1	1.6	1.1
T_2	1.6	1.2
$p\ (T_0\text{-}T_1)^a$	***0.015***	***0.0003***
$p\ (T_0\text{-}T_2)^a$	***0.005***	***0.001***
$p\ (T_0\text{-}T_1\text{-}T_2)^b$	***0.018***	***0.00002***

Group A: only pharmacological prophylaxis; Group B: pharmacological prophylaxis and acupuncture. [a] Wilcoxon Test – not parametric for paired samples; [b] Friedman Test – not parametric for repeated samples. T_0=baseline; T_1= at 2-month follow up; T_2= at 4-month follow up

Table 3. Effect of acupuncture on selected outcomes in the overall headache (any) study population: comparison between Group A and Group B at the study baseline, 2-month and 4-month follow up, respectively.

	Group A	Group B
HDI (median)		
T_0	3.0	4.0
T_1	2.5	3.0
T_2	2.5	2.0
$p\ (T_0\text{-}T_1)^a$	***0.026***	***0.008***
$p\ (T_0\text{-}T_1\text{-}T_2)^b$	***0.021***	***<0.0001***
BDI2 (median)		
T_0	2.0	1.0
T_1	1.0	1.0
T_2	1.0	1.0
$p\ (T_0\text{-}T_1)^a$	***0.033***	***0.022***
$p\ (T_0\text{-}T_1\text{-}T_2)^b$	***0.005***	*0.091*
MIDAS (median)		
T_0	-	-
T_1	-	-
T_2	3.0	3.0
$p\ (T_0\text{-}T_1)^a$	-	-
$p\ (T_0\text{-}T_1\text{-}T_2)^b$	*0.705*	***0.002***
STAI-Trait (median)		
T_0	45.0	45.0
T_1	43.0	42.5
T_2	44.0	39.0
$p\ (T_0\text{-}T_1)^a$	***0.005***	***0.026***
$p\ (T_0\text{-}T_1\text{-}T_2)^b$	***0.017***	***0.020***
SF-36 – Physical functioning (median)		
T_0	92.5	90.0
T_1	90.0	90.0
T_2	90.0	90.0
$p\ (T_0\text{-}T_1)^a$	*0.452*	*0.888*
$p\ (T_0\text{-}T_1\text{-}T_2)^b$	*0.233*	*0.180*
SF-36 – Limitations due to physical functioning (median)		
T_0	*0.0*	*25.0*
T_1	*50.0*	*75.0*
T_2	*25.0*	*100*
$p\ (T_0\text{-}T_1)^a$	*0.057*	***0.005***
$p\ (T_0\text{-}T_1\text{-}T_2)^b$	*0.211*	***0.020***
SF-36 – Limitations due to emotional problems (median)		
T_0	0.0	83.4
T_1	83.5	100

Table 3. Effect of acupuncture on selected outcomes in the overall headache (any) study population: comparison between Group A and Group B at the study baseline, 2-month and 4-month follow up, respectively.

	Group A	Group B
SF-36 – Limitations due to emotional problems (median)		
T_2	100	100
$p\ (T_0\text{-}T_1)^a$	***0.005***	*0.077*
$p\ (T_0\text{-}T_1\text{-}T_2)^b$	***0.001***	*0.247*
SF-36 – Vitality (median)		
T_0	*45.0*	*50.0*
T_1	*55.0*	*52.5*
T_2	*55.0*	*52.5*
$p\ (T_0\text{-}T_1)^a$	***0.004***	*0.080*
$p\ (T_0\text{-}T_1\text{-}T_2)^b$	***0.011***	*0.158*
SF-36 – Emotional well-being (median)		
T_0	52.0	68.0
T_1	72.0	64.0
T_2	72.0	72.0
$p\ (T_0\text{-}T_1)^a$	***0.0005***	*0.484*
$p\ (T_0\text{-}T_1\text{-}T_2)^b$	***0.0001***	*0.191*
SF-36 – Bodily pain (median)		
T_0	*45.0*	*45.0*
T_1	*51.3*	*66.3*
T_2	*57.5*	*62.5*
$p\ (T_0\text{-}T_1)^a$	***0.047***	***0.003***
$p\ (T_0\text{-}T_1\text{-}T_2)^b$	***0.013***	***0.007***
SF-36 – Social functioning (median)		
T_0	*62.5*	*50.0*
T_1	*75.0*	*75.0*
T_2	*75.0*	*75.0*
$p\ (T_0\text{-}T_1)^a$	*0.437*	*0.056*
$p\ (T_0\text{-}T_1\text{-}T_2)^b$	*0.133*	*0.106*
SF-36 – General health (median)		
T_0	*40.0*	*57.5*
T_1	*37.5*	*60.0*
T_2	*35.5*	*57.5*
$p\ (T_0\text{-}T_1)^a$	*0.372*	*0.895*
$p\ (T_0\text{-}T_1\text{-}T_2)^b$	*0.556*	*0.495*

Group A: only pharmacological prophylaxis; Group B: pharmacological prophylaxis and acupuncture. HDI= Henry Ford Headache Disability Inventory - beta; BDI2= Beck Depression Inventory-II; MIDAS= Migraine Disability Assessment; STAI= State-Trait Anxiety Inventory; SF-36= Short Form 36 Health Survey. [a] Wilcoxon Test – not parametric for paired samples; [b] Friedman Test – not parametric for repeated samples. T_0=baseline; T_1= at 2-month follow up; T_2= at 4-month follow up

Table 4. Effect of acupuncture on selected outcomes in the study population with tension-type headache only: comparison between Group A and Group B at the study baseline, 2-month and 4-month follow up, respectively.

	Group A	Group B
HDI (median)		
T_0	3.0	3.0
T_1	3.0	2.0
T_2	3.0	2.0
$p\ (T_0\text{-}T_1)^a$	*0.317*	*0.096*
$p\ (T_0\text{-}T_1\text{-}T_2)^b$	*0.174*	*0.076*
BDI2 (median)		
T_0	2.0	2.0
T_1	1.5	1.0
T_2	1.5	1.0
$p\ (T_0\text{-}T_1)^a$	***0.025***	*0.059*
$p\ (T_0\text{-}T_1\text{-}T_2)^b$	***0.040***	*0.074*
MIDAS (median)		
T_0	-	-
T_1	-	-
T_2	3.5	2.0
$p\ (T_0\text{-}T_1)^a$	-	-
$p\ (T_0\text{-}T_1\text{-}T_2)^b$	*0.564*	***0.046***
STAI-Trait (median)		
T_0	54.0	40.0
T_1	52.0	39.0
T_2	50.0	36.0
$p\ (T_0\text{-}T_1)^a$	*0.156*	*0.235*
$p\ (T_0\text{-}T_1\text{-}T_2)^b$	***0.023***	*0.121*
SF-36 – Physical functioning (median)		
T_0	95.0	90.0
T_1	90.0	95.0
T_2	95.0	100
$p\ (T_0\text{-}T_1)^a$	*0.157*	*0.104*
$p\ (T_0\text{-}T_1\text{-}T_2)^b$	*0.135*	***0.016***
SF-36 – Limitations due to physical functioning (median)		
T_0	*0.0*	*25.0*
T_1	*37.5*	*100*
T_2	*0.0*	*100*
$p\ (T_0\text{-}T_1)^a$	*0.257*	*0.058*
$p\ (T_0\text{-}T_1\text{-}T_2)^b$	*0.268*	*0.196*
SF-36 – Limitations due to emotional problems (median)		
T_0	0.0	100
T_1	83.4	100
T_2	100	66.7

Table 4. Effect of acupuncture on selected outcomes in the study population with tension-type headache only: comparison between Group A and Group B at the study baseline, 2-month and 4-month follow up, respectively.

	Group A	Group B
SF-36 – Limitations due to emotional problems (median)		
***p* (T_0-T_1)[a]**	***0.026***	*.7050*
***p* (T_0-T_1-T_2)[b]**	***0.028***	*0.309*
SF-36 – Vitality (median)		
T_0	40.0	55.0
T_1	36.3	60.0
T_2	52.5	55.0
***p* (T_0-T_1)[a]**	*0.256*	*0.201*
***p* (T_0-T_1-T_2)[b]**	*0.177*	*0.482*
SF-36 – Emotional well-being (median)		
T_0	38.0	72.0
T_1	58.0	72.0
T_2	64.0	76.0
***p* (T_0-T_1)[a]**	***0.011***	*0.932*
***p* (T_0-T_1-T_2)[b]**	***0.002***	*0.887*
SF-36 – Bodily pain (median)		
T_0	45.0	45.0
T_1	45.0	77.5
T_2	56.2	80.0
***p* (T_0-T_1)[a]**	*0.269*	***0.042***
***p* (T_0-T_1-T_2)[b]**	*0.066*	***0.022***
SF-36 – Social functioning (median)		
T_0	95.0	62.5
T_1	62.5	87.5
T_2	75.0	87.5
***p* (T_0-T_1)[a]**	*0.516*	*0.131*
***p* (T_0-T_1-T_2)[b]**	*0.459*	*0.368*
SF-36 – General health (median)		
T_0	*32.5*	*70.0*
T_1	*35.0*	*80.0*
T_2	*32.5*	*85.0*
***p* (T_0-T_1)[a]**	*0.679*	*0.168*
***p* (T_0-T_1-T_2)[b]**	*0.607*	*0.152*

Group A: only pharmacological prophylaxis; Group B: pharmacological prophylaxis and acupuncture; HDI= Henry Ford Headache Disability Inventory - beta; BDI2= Beck Depression Inventory-II; MIDAS= Migraine Disability Assessment; STAI= State-Trait Anxiety Inventory; SF-36= Short Form 36 Health Survey; [a] Wilcoxon Test – not parametric for paired samples; [b] Friedman Test – not parametric for repeated samples; T_0=baseline; T_1= at 2-month follow up; T_2= at 4-month follow up

effects it entails (31, 32). To evaluate its prophylactic efficacy, a drug requires at least two months at a target dosage (33), a dosage that is achieved on average in no less than one month, if the dosage includes a gradual increase. Drug prophylaxis has been seen to take up to one or two months before any beneficial effect is achieved. Based on the many studies demonstrating the short-term effectiveness of acupuncture on pain, the intervention perspective of the present study mainly focused on this time interval (34). Many studies foresee cycles of at least six sessions per week; cycles that therefore last a minimum of two months. Weekly frequency is usually the most common option but there is evidence of efficacy even for bi-weekly or alternate day protocols (13,35). These considerations led us to prepare a non-prolonged, but potentially effective, treatment with acupuncture concentrated in one month with bi-weekly sessions. The choice of assessment methods aimed, as in other studies, at considering both the specifics of the disorder (data from the headache diary) and its implications on the patient. In the case of headache, the main purpose is to combat the disability that it entails, the success of which depends on the treatment proposed which can determine the patient's perception, expectations and satisfaction. As far as the results of the study are concerned, assignment to alternating consecutive series made it possible to obtain two homogeneous groups, with well-distributed variables and no significant differences. The totality of the sample was migraine, with a proportion of subjects presenting, in addition, a coexistence of tension-type headache (Group A: eight; Group B: seven), whose primary outcomes were evaluated separately in a subanalysis. The dominance of the migraine pathology is understandable in relation to the greater disability it implies and, therefore, the indication for prophylactic therapy; in fact, it is less likely that a frequent episodic tension-type headache alone leads to a significant disability. This result was partially confirmed in the subanalysis that compared, at baseline across the entire sample, all migraine patients (chronic and non-chronic forms) with mixed headache patients (migraine and tension) and further indicated that disability due to headache (HDI score) was higher in percentage terms in migraine patients (severe grade) than in mixed headaches (moderate grade). Data from the hea-

Table 5: Impact of acupuncture on the main clinical outcomes (median) in the study population affected by mixed headache with migraine and tension components

	T0	T1	*p*[a]	T0	T1	T2	*p*[a]
HDI							
(A) Pharmacoprophylaxis	3.0	3.0	*0.317*	3.0	3.0	3.0	*0.174*
(B) Pharm+Acupuncture	3.0	2.0	*0.096*	3.0	3.0	2.0	*0.076*
BDI2							
(A) Pharmacoprophylaxis	2.0	1.5	***0.025***	2.0	1.5	1.5	***0.040***
(B) Pharm+Acupuncture	2.0	1.0	*0.059*	2.0	1.0	1.0	*0.074*
MIDAS							
(A) Pharmacoprophylaxis	-	-		3.0	-	3.5	*0.564*
(B) Pharm+Acupuncture	-	-		4.0	-	2.0	***0.046***
STAI-Trait							
(A) Pharmacoprophylaxis	54.0	52.0	*0.156*	54.0	50.5	50.0	***0.023***
(B) Pharm+Acupuncture	40.0	39.0	*0.235*	40.0	36.0	36.0	*0.121*
SF-36							
Physical function							
(A) Pharmacoprophylaxis	95.0	95.0	*0.157*	95.0	95.0	95.0	*0.135*
(B) Pharm+Acupuncture	90.0	95.0	*0.104*	90.0	95.0	100	***0.016***
Limitations due to physical function							
(A) Pharmacoprophylaxis	0.0	37.5	*0.257*	0.0	37.5	0.0	*0.268*
(B) Pharm+Acupuncture	25.0	100	*0.058*	25.0	100	100	*0.196*
Limitations due to emotional problems							
(A) Pharmacoprophylaxis	0.0	83.4	***0.026***	0.0	83.4	100	***0.028***
(B) Pharm+Acupuncture	100	100	*0.705*	100	100	66.7	*0.309*
Activity							
(A) Pharmacoprophylaxis	40.0	36.3	*0.256*	40.0	47.5	52.5	*0.177*
(B) Pharm+Acupuncture	55.0	60.0	*0.201*	55.0	60.0	55.0	*0.482*
Emotional well-being							
(A) Pharmacoprophylaxis	38.0	58.0	***0.011***	48.0	72.0	64.0	***0.002***
(B) Pharm+Acupuncture	72.0	72.0	*0.932*	72.0	72.0	76.0	*0.887*
Pain							
(A) Pharmacoprophylaxis	45.0	45.0	*0.269*	45.0	50.0	56.2	*0.066*
(B) Pharm+Acupuncture	45.0	77.5	***0.042***	45.0	77.5	80.0	***0.022***
Social functioning							
(A) Pharmacoprophylaxis	95.0	62.5	*0.516*	62.5	62.5	75.0	*0.459*
(B) Pharm+Acupuncture	62.5	87.5	*0.131*	62.5	87.5	87.5	*0.368*
General health							
(A) Pharmacoprophylaxis	32.5	35.0	*0.679*	32.5	35.0	32.5	*0.607*
(B) Pharm+Acupuncture	70.0	80.0	*0.168*	70.0	80.0	85.0	*0.152*

[a] Wilcoxon Test; [b] Friedman Test; T_0=baseline; T_1= at 2-month follow up; T_2= at 4-month follow up; HDI= Henry Ford Headache Disability Inventory - beta; BDI2= Beck Depression Inventory-II; MIDAS= Migraine Disability Assessment; STAI= State-Trait Anxiety Inventory; SF-36= Short Form 36 Health Survey

dache diaries showed that both acupuncture and drug prophylaxis determined a positive trend in the primary outcomes for migraine, but acupuncture reports greater and more persistent effects. Two months after the start of acupuncture treatments, Group B recorded significant improvements in all items: average intensity of attacks, disability, frequency, intake of analgesics and triptans; while in Group A, the significance was minimal and only for frequency and disability. At the fourth month, these improvements were maintained with acupuncture, but less prominent than in the short term (two months). In Group A, the delayed improvements in disability, in frequency and the intake of analgesics, confirmed the dynamics of the effect of pharmacoprophylaxis. Unlike acupuncture, drug prophylaxis did not result in significant improvements in terms of the intensity and quantity of triptans taken. Headache-specific secondary outcomes (in which the tension component of patients with mixed headache is included) showed improvements in line with the primary ones. Perceived disability was greatly reduced in patients in Group B, at both 2nd and 4th months, with a positive trend passing from severe to mild (HDI) and from severe to moderate (MIDAS), respectively. For anxiety trait and depressive symptoms, there were no notable differences between the two groups.

The sub-analysis between patients with single migraine headache vs mixed headache provides interesting data especially if we consider that the distinction between them cannot be clearly seen on the basis of the pathology. It may be observed that the group of patients with mixed headache was on average ten years younger than migraine sufferers and that, out of 15, eight were non-manual workers and five were students. As already mentioned, the perceived disability of patients with mixed headache was on average less than migraine sufferers, and 60% had symptoms of mild depression, an association that is reflected in other studies (36, 37, 38). In the headache diaries, the ability to separately characterize migraine attacks from tension-type attacks required a specific evaluation. Despite the small numbers of the sample with mixed headache, the tension component tended to highlight a greater benefit associated with acupuncture in the primary outcomes. In the second month, a significant reduction was noticed only in frequency, although the impact on disability and average intensity was better than in the group without acupuncture. At the 4th month, as already seen for migraine, pharmacological prophylaxis showed its effects, even if minor and not significant compared to those of Group B. In the timeline, the effectiveness of acupuncture in reducing frequency and disability of the disorder was greater. Acupuncture was also effective on tension-type headaches, although with a lesser effect than on migraine, as is confirmed in the literature (13). The impact of acupuncture on the clinical outcomes of patients with mixed headache reflected the results seen in the whole sample with less impact (in fact, it was not possible to evaluate the impact of the tension component alone in these data). The effects of acupuncture on perceived disability were significantly better in the 2nd month of treatment from moderate to mild in the HDI test, and in the 4th month from severe to mild in MIDAS. Regarding anxiety trait and depressive symptoms, in Group B there were improvements (but less significant compared to Group A). Acupuncture brought improvements to the scales of physical function and pain perception, the latter visible already in the 2nd month of treatment. The results of the health status questionnaire indicate that acupuncture led to improvements on the scales of physical limitation and perception of pain, whereas pharmacoprophylaxis was more effective on scales inherent to the emotional sphere and energy levels. Pharmacoprophylaxis, in the entire sample, led to better effects on emotional well-being and fewer limitations due to emotional problems.

The high prevalence of headache together with the not very restrictive criteria of this study (indication of pharmacological prophylaxis for migraine and/or tension-type headache) has made it possible to select a sample that is representative of "real life" among patients with headaches. The participants in this study are adults who commonly suffer from headache-related disability and who would mostly benefit from a promptly effective therapy. Acupuncture, thanks to the almost total absence of side effects and its accessibility, could meet these needs by improving the effects of pharmacological prophylaxis. The expectations and satisfaction provided by the therapy are subjective parameters that affect the definition of an effective therapy. The short course of therapy and the long observation

times vis-a-vis, exclude the possible interference of a placebo effect. The operator-dependence of treatment with acupuncture (only one acupuncturist) and the preparation of the standardized therapeutic protocol on a symptomatic basis, may alter and contain possible distortions in the outcomes. Usual care appears to be the best way to test the effectiveness of an intervention with acupuncture, given that sham is a very similar practice, therefore considered not inert (39, 40). According to the literature, however, compared to normal acupuncture, sham has lower and less lasting significance for migraine and chronic migraine and none for tension-type headache (12, 13).

The planning process of this pilot study encountered some problems that may be described, by focusing on the considerations and strategies put in place to overcome them. The number of participants already poses a limit in the preparatory phase. The three recruitment centers have a limited catchment area, mainly in the province of Ferrara and some neighboring municipalities in the provinces of Rovigo and Bologna, and the only dedicated center (Ferrara Headache Center) is active just two days a month. This problem could be dealt with by opting for not very restrictive inclusion criteria, which still however, aim at selecting a specific target: patients with disabling headaches serious enough to justify drug prophylaxis (13). This factor, together with the relatively limited duration of the recruitment period (one year) and the necessary exclusion criteria, undoubtedly weigh on the size of the sample. In this context a second limit exists: the recruitment was carried out by assignment to alternating consecutive series in the two groups. The lack of comparison with a sham treatment constitutes a further limit.

The data obtained from this pilot study confirm that acupuncture, as a supportive therapy for drug prophylaxis for migraine and tension-type headache, confers greater benefits than drug prophylaxis alone. This improvement particularly affects the average intensity of attacks, frequency (days / month), monthly doses of analgesics and triptans taken, but above all the disability related to the attacks. The improvements are noticed in the medium term (four months) and even more in the short term (two months). The normal times required to evaluate the effects of a drug prophylaxis are at least three-four months and these results are seldom prompt, unlike the possible side effects, thus an add-on acupuncture intervention, even if concentrated in a month, as in this case, can represent an excellent supplement to drugs, its effects covering the first months of prophylaxis, often crucial for the patient's therapeutic compliance. The potential advantages of an integrated approach for the patient would therefore result in better prophylactic efficacy and a higher adherence to therapy, with a lower risk of abandonment in the first months, where the balance of "benefits / side effects" of pharmacoprophylaxis may tend towards the latter. Although the study lacks observations on long-term effects, the data collected so far offer good prospects for further study, including large scale cohorts.

References

1. Packard RC. What does the headache patient want?. Headache. 1979;19(7):370–374.
2. Jensen R, Stovner LJ. Epidemiology and comorbidity of headache. Lancet Neurol. 2008;7(4):354–361.
3. Saylor D, Steiner TJ. The Global Burden of Headache. Semin Neurol. 2018;38(2):182–190.
4. Stovner LJ, Andree C. Prevalence of headache in Europe: a review for the Eurolight project. J Headache Pain. 2010;11(4):289–299.
5. Linde M, Gustavsson A, Stovner LJ, et al. The cost of headache disorders in Europe: the Eurolight project. Eur J Neurol. 2012;19(5):703–711.
6. D'Amico D, Grazzi L, Curone M, Leonardi M, Raggi A. Cost of medication overuse headache in Italian patients at the time-point of withdrawal: a retrospective study based on real data. Neurol Sci. 2017;38(Suppl 1):3–6.
7. Rasmussen BK. Epidemiology of headache. Cephalalgia. 1995;15(1):45–68.
8. Delaruelle Z, Ivanova TA, Khan S, et al. Male and female sex hormones in primary headaches. J Headache Pain. 2018;19(1):117. Published 2018 Nov 29
9. Yang ES, Li PW, Nilius B, Li G. Ancient Chinese medicine and mechanistic evidence of acupuncture physiology. Pflugers Arch. 2011 Nov;462(5):645-53. doi: 10.1007/s00424-011-1017-3. Epub 2011 Aug 26. PMID: 21870056; PMCID: PMC3192271.
10. Zhou W, Benharash P. Effects and mechanisms of acupuncture based on the principle of meridians. J Acupunct Meridian Stud. 2014 Aug;7(4):190-3. doi: 10.1016/j.jams.2014.02.007. Epub 2014 Jun 24. PMID: 25151452.
11. Wojcikowski K, Vigar VJ, Oliver CJ. New Concepts of Chronic Pain and the Potential Role of Complementary Therapies. Altern Ther Health Med. 2020 Feb;26(S1):18-31. PMID: 29428928.

12. Vickers AJ, Cronin AM, Maschino AC, et al. Acupuncture for chronic pain: individual patient data meta-analysis. Arch Intern Med. 2012;172(19):1444–1453.
13. Linde K, Allais G, Brinkhaus B, et al. Acupuncture for the prevention of episodic migraine. Cochrane Database Syst Rev. 2016;2016(6):CD001218. Published 2016 Jun 28.
14. Giannini, G. & Nicodemo, Marianna & Favoni, Valentina & Matra, A. & Giovanardi, C. & Pierangeli, G. & Cortelli, Pietro & Cevoli, Sabina. (2015). The acumigran study: a randomized controlled clinical trial on the efficacy of acupuncture for migraine prophylaxis. Cephalalgia. 35. 34-34.
15. Coeytaux RR, Befus D. Role of Acupuncture in the Treatment or Prevention of Migraine, Tension-Type Headache, or Chronic Headache Disorders. Headache. 2016;56(7):1238–1240.
16. Gasser PA. Creating a headache diary. J Am Acad Nurse Pract. 1991;3(1):53–55.
17. Downie WW, Leatham PA, Rhind VM, Wright V, Branco JA, Anderson JA. Studies with pain rating scales. Ann Rheum Dis. 1978;37(4):378–381.
18. Nappi G, Jensen R, Nappi RE, Sances G, Torelli P, Olesen J. Diaries and calendars for migraine. A review. Cephalalgia. 2006;26(8):905–916.
19. Torelli P, Jensen R. Headache diaries and calendars. Handb Clin Neurol. 2010;97:137–146.
20. Rossi P, Di Lorenzo G, Malpezzi MG, et al. Depressive symptoms and insecure attachment as predictors of disability in a clinical population of patients with episodic and chronic migraine. Headache. 2005;45(5):561–570.
21. Santangelo G, Russo A, Trojano L, et al. Cognitive dysfunctions and psychological symptoms in migraine without aura: a cross-sectional study. J Headache Pain. 2016;17(1):76.
22. Jacobson GP, Ramadan NM, Aggarwal SK, Newman CW. The Henry Ford Hospital Headache Disability Inventory (HDI). Neurology. 1994;44(5):837–842.
23. Jacobson GP, Ramadan NM, Norris L, Newman CW. Headache disability inventory (HDI): short-term test-retest reliability and spouse perceptions. Headache. 1995;35(9):534–539.
24. Stewart WF, Lipton RB, Kolodner K, Liberman J, Sawyer J. Reliability of the migraine disability assessment score in a population-based sample of headache sufferers. Cephalalgia. 1999;19(2):107–74.
25. Stewart WF, Lipton RB, Dowson AJ, Sawyer J. Development and testing of the Migraine Disability Assessment (MIDAS) Questionnaire to assess headache-related disability. Neurology. 2001;56(6 Suppl 1):S20–S28.
26. Julian LJ. Measures of anxiety: State-Trait Anxiety Inventory (STAI), Beck Anxiety Inventory (BAI), and Hospital Anxiety and Depression Scale-Anxiety (HADS-A). Arthritis Care Res (Hoboken). 2011;63 Suppl 11(0 11):S467–S472.
27. Rizzo A, Muscatello MRA, Autunno M, et al. Le emozioni negative nei soggetti cefalalgici [Negative emotions in headache patients.]. Recenti Prog Med. 2018;109(7):393–397.
28. Jackson-Koku G. Beck Depression Inventory. Occup Med (Lond). 2016;66(2):174–175.
29. Apolone G, Mosconi P. The Italian SF-36 Health Survey: translation, validation and norming. J Clin Epidemiol. 1998;51(11):1025–1036.
30. Brazier JE, Harper R, Jones NM, et al. Validating the SF-36 health survey questionnaire: new outcome measure for primary care. BMJ. 1992;305(6846):160–164.
31. Silberstein SD, Goadsby PJ. Migraine: preventive treatment. Cephalalgia. 2002;22(7):491–512.
32. Young WB, Hopkins MM, Shechter AL, Silberstein SD. Topiramate: a case series study in migraine prophylaxis. Cephalalgia. 2002;22(8):659–663.
33. Loder E, Rizzoli P. Pharmacologic Prevention of Migraine: A Narrative Review of the State of the Art in 2018. Headache. 2018;58 Suppl 3:218–229.
34. Endres HG, Diener HC, Molsberger A. Role of acupuncture in the treatment of migraine. Expert Rev Neurother. 2007;7(9):1121–1134.
35. Zhao L, Chen J, Li Y, et al. The Long-term Effect of Acupuncture for Migraine Prophylaxis: A Randomized Clinical Trial. JAMA Intern Med. 2017;177(4):508–515.
36. Fuensalida-Novo S, Palacios-Ceña M, Fernández-Muñoz JJ, et al. The burden of headache is associated to pain interference, depression and headache duration in chronic tension type headache: a 1-year longitudinal study. J Headache Pain. 2017;18(1):119. Published 2017 Dec 28.
37. Ashina S, Bendtsen L, Buse DC, Lyngberg AC, Lipton RB, Jensen R. Neuroticism, depression and pain perception in migraine and tension-type headache. Acta Neurol Scand. 2017;136(5):470–476.
38. Palacios-Ceña M, Fernández-Muñoz JJ, Castaldo M, et al. The association of headache frequency with pain interference and the burden of disease is mediated by depression and sleep quality, but not anxiety, in chronic tension type headache. J Headache Pain. 2017;18(1):19.
39. MacPherson H, Vertosick E, Lewith G, et al. Influence of control group on effect size in trials of acupuncture for chronic pain: a secondary analysis of an individual patient data meta-analysis. PLoS One. 2014;9(4):e93739. Published 2014 Apr 4.
40. Birch S. A review and analysis of placebo treatments, placebo effects, and placebo controls in trials of medical procedures when sham is not inert. J Altern Complement Med. 2006;12(3):303–310.