

**IDENTIFICATION AND CHARACTERIZATION OF MUSCARINIC RECEPTORS IN HUMAN MALE DETRUSOR AND IN PRIMARY CELL CULTURE OF DETRUSOR LEIOMYOCYTES**

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**INTRODUCTION AND AIM OF THE STUDY** Activation of the parasympathetic cholinergic system is the major pathway by which bladder contraction is achieved in humans, both by direct stimulation and by inhibiting noradrenergic-induced relaxation. The postjunctional effects of acetylcholine (ACh) are induced via muscarinic receptors, that are widely expressed, both at the messenger RNA (mRNA) and protein levels in urinary bladder smooth muscle. Aim of this study is, firstly, to characterize the muscarinic receptor subtypes expressed in the human detrusor muscle, investigating whether there are significant age-dependent differences. Then, we established primary culture of cells derived from human detrusor to study the expression of muscarinic receptors in a pure cell population. The availability of these cell cultures will help to better understand the role of postsynaptic muscarinic receptors in human detrusor.

**MATERIALS AND METHODS Patients.** The study was carried out on 26 men, divided, based on age, in: men < 65 years (yr) (adult, n=10; 61 ± 3,3 yr); men > 65 yr (old, n=16; 73 ± 6,8 yr). Informed consent was obtained from all subjects. Tissues were obtained as discarded tissues after radical cystectomy for bladder cancer. All patients displayed a normal micturition frequency, with no symptoms of urgency or obstruction. Samples (approximately 90-100 mg), which were not macroscopically invaded by tumor, were taken from the bladder dome, cut into two

pieces. One of the pieces was used for the histological detection of tumor, whereas for the other, the detrusor was separated from the mucosa by microdissection, cut into three pieces and then immediately frozen in liquid nitrogen within 30 minutes and used for this study. For the establishment of leiomyocytes culture, a sample of fresh detrusor was placed in a M199 (45%)-DMEM (45%)- serum (10%) medium and used for the cell culture

described below. Experiments were carried out once the area was diagnosed as tumor-free. **Q-RT-PCR.** The same amount of tissue for each subject was used to arrange three different pools for every group of patients (3 pools were made of different samples from adult men and 3 from samples from old men; preliminary experiments demonstrated the superimposable muscarinic receptor gene expression between pools). RNA extraction, cDNA transcription and quantitative RT-PCR (Q-RT-PCR) were performed as described in Arrighi et al<sup>1</sup>. Muscarinic receptor expression was evaluated by Q-RT-PCR, using SYBRGreen as fluorochrome and the  $\beta$  actin as reference gene. The threshold cycle (Ct) values for the amplicons of the gene studied was calculated. **Western blot.** The same procedure above described was followed to combine pools of tissues in order to analyze protein expression by western blot. Equal amounts of protein (50  $\mu$ g) were separated by electrophoresis and electroblotted to a nitrocellulose membrane. The protein detection was conducted as described in Arrighi et al<sup>1</sup>. **Radioligand binding.** Experiments were performed according to Goepel et al<sup>2</sup>, with minimal modifications. The kinetic parameters, Kd and Bmax were calculated by nonlinear regression of saturation curves and Scatchard analysis, using the GraphPad PRISM 4 software. Results are reported as mean  $\pm$  SE of four different experiments run in triplicate. **Primary cell culture.** Samples of detrusor muscle were cut in fragments (about 1 mm<sup>3</sup>) and kept in culture in M199 (45%)-DMEM (45%)-serum (10%) medium.

The muscle origin of cells was demonstrated by the staining with  $\alpha$ -actin specific antibodies. RNA extraction, cDNA transcription and quantitative RT-PCR (Q-RT-PCR) were performed as described in Arrighi et al<sup>1</sup>.

## RESULTS.

Analysis of saturation binding curves confirmed that muscarinic receptors are expressed in the detrusor: the Bmax for the adult men detrusor samples was indeed  $77,6 \pm 3,5$  fmol/mg prot, while a lower density, although not statistically significant, was observed in the detrusor of old men ( $64 \pm 2,4$  fmol/mg). The calculated Kd showed no significant differences:  $0,11 \pm 0,27$  nM in adult men,  $0,10 \pm 0,21$  nM in old men, consistent with those obtained for cloned receptors.

Q-RT-PCR demonstrated that each muscarinic receptor subtype was expressed in human detrusor. The molecular expression of each subtype of the M<sub>1</sub> receptor family was observed and it was not influenced by age. M<sub>2</sub> receptor family transcripts revealed that both M<sub>2</sub> and M<sub>4</sub> were detected and we could observe an age-related modification of the M<sub>4</sub> transcripts; indeed, M<sub>4</sub> mRNA was lower in old than in adult men ( $p < 0.05$ ), but

higher in old than in adult women ( $p < 0.05$ ). The western blot followed by quantification confirmed that the mRNAs were translated into proteins, maintaining the range of expression observed at mRNA level:  $M_2 > M_3 > M_4 > M_1 > M_5$ .

In primary culture of leiomyocytes (passage 2), the molecular detection of muscarinic receptors by Q-RT-PCR revealed that, according to the results obtained in biopsies, each subtype is expressed, with a high expression of the  $M_2$  subtype.

## **DISCUSSION AND CONCLUSION.**

Muscarinic receptors are present in human detrusor, both at the mRNA and protein level. In particular, according to literature, the  $M_2$  and  $M_3$  subtypes are more expressed than  $M_4$  and  $M_1$ ,  $M_5$  showed a very weak expression.

The selective reduction of  $M_4$  receptor mRNA observed in old men indicates that this subtype could play an active role in the pathophysiology of micturition and in the age-related changes in bladder function. The reduced expression of the  $M_4$  may also account for the reduced (although not significant)  $B_{max}$  in the radioligand binding observed in old men compared to adult ones. Protein analyses partially confirm Q-RT-PCR results. According to literature,  $M_2$  and  $M_3$  subtypes are expressed at higher level compared to the other subtypes.

The establishment of primary cell cultures of human leiomyocytes and the results obtained by studying the muscarinic receptor expression pointed out that this approach may represent a suitable model to study the biology and the physiology of detrusor at cellular level and it may aid to the better understanding of the interaction between different receptors.

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**ROLE OF PRE-OPERATIVE URODYNAMICS IN WOMEN WITH CLINICAL DIAGNOSIS OF STRESS URINARY INCONTINENCE.**

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**INTRODUCTION AND AIM OF THE STUDY**

Pre-operative full urodynamic evaluation in women with clinical diagnosis of Genuine Stress Urinary Incontinence was not considered recommended in recent reports as a routine practice (1). Aim of the study was to compare our data with other reports in literature on this field with particular regard to the sensitivity and specificity of “simple” clinical diagnosis and, consequently, to contribute in the debate on the role and significance of the urodynamic examinations prior to the continence surgical procedures.

**MATERIALS AND METHODS**

A retrospective analysis of 278 full urodynamic records regarding a group of female patients referred to our Unit for a pre-operative assessment of Stress Urinary Incontinence between January 2006 and June 2007, was conducted on the basis of designed selective criteria (Table 1). Data processing included patient’s age, history and clinical diagnosis and urodynamic evidences of 195 subjects aged between 33 and 80 years. All the patients achieved a clinical diagnosis of Stress Urinary Incontinence (SUI) that was compared to the reports of full urodynamic examination (including free uroflowmetries, water cystometry and perfusional urethral pressure profile or pressure-flow study).

Table I. Exclusion Criteria

|                             |                                      |
|-----------------------------|--------------------------------------|
| Overactive Bladder Symptoms | Neurogenic Diseases                  |
| Urinary Tract Infection     | Previous Pelvic / Continence Surgery |

## RESULTS

All records regarded patients sent to our Unit by specialists (56% gynaecologists, 44% urologists). 144 patients among the 195 selected (73.8%) were registered with a diagnosis of isolate Urodynamic Stress Incontinence (USI). Thirty-eight patients (19.4%) had a negative report (No USI, No Urodynamic Abnormality) while 13 patients (6.6%) presented different urodynamic reports of unbalanced void with or without Urodynamic Urinary Incontinence. The diagnosis registered showed Detrusor Overactivity (4 pz), Detrusor Underactivity with Sphincteric Weakness (6 pz) and Detrusor Impaired Contractility (3 pz). Sensitivity of the clinical diagnosis of SUI was 19% while Specificity was 80%. The epidemiological evaluation showed a Positive Predictive Value of 74% and Negative Predictive Value of 73%. In Table 2 we have been plotted our data versus other reports (1, 2).

| Test        | Our Sample | Bristol 2007 | NICE 2006    |
|-------------|------------|--------------|--------------|
| Sensitivity | 19%        | 11%          | 66%(17-83%)  |
| Specificity | 80%        | 98%          | 83% (49-92%) |
| PPV         | 74%        | 74%          | 70%(41-95%)  |
| NPV         | 73%        | 72%          | 69%(49-65%)  |

## DISCUSSION

Our data showed only light differences in comparison with the Bristol reports (2) but in our experience a low sensitivity and a less high specificity of clinical diagnosis in the sample processed could be due to the selection criteria adopted. Nevertheless, even our sample revealed a 26% of different urodynamic diagnoses that could become inappropriate to the surgical procedure indication or liable to bad results or adverse effects on the outcome of our patients.

## CONCLUSION

Even in our experience, an accurate and complete urodynamic evaluation remains a useful tool also when clinical diagnosis of SUI seems to be sufficient for surgical correction. In our opinion, further data reviews and statistical comparison between

different experiences cannot represent the key to establish a role of urodynamics that remains absolutely recommended in pre-operative assessment of SUI.

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**PERFORMANCE OF INCONTINENCE IMPACT QUESTIONNAIRE (IIQ) IN ITALIAN POPULATION ACCORDING TO EDUCATIONAL STATUS**

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**INTRODUCTION AND AIM OF THE STUDY**

Symptoms correlated with pelvic organ prolapse (POP) and/or urinary incontinence (UI) are frequent and disabling. The assessment of distress due to such symptoms, when women carrying out daily activities, is however very difficult to achieve because distress is a subjective parameter which is often dissociated from the severity of symptoms but linked to individual emotional and psychosocial factors. As the patient's perception of her disturbances seems to be a major factor influencing her quality of life (QoL), several questionnaires have been developed to assess QoL scores. Many self-administered questionnaires identify and assess the severity of symptoms and the extent of their impact on routine activities of daily life and patients related outcome after a therapeutic procedure [1, 2]. Impact Incontinence Quality of life (IIQ) was the most frequently used measure of incontinence-specific QoL in research studies and outcome assessment after disease oriented therapy. It assesses the social, psychological and physical well-being of the patient and the impact of the uro-gynaecological pathology on her daily life. It has proved to be very reproducible, with high sensitivity for the patient's psycho-social condition even though it was originally developed using a selected population of highly educated women over 45 years of age. To the best of our knowledge, no study investigated the performance of IIQ on Italian population. Aim of this study is to determine whether the IIQ questionnaire is suitable for Italian patients in terms of comprehension, difficulty in answering questions and relation with the patient's underlying uro-genital pathology.

**MATERIALS AND METHODS**

The study prospectively recruited 172 consecutive patients (mean age  $61 \pm 10$  years; range 27–90) from January 2005 to December 2006, who were referred to a single tertiary urban teaching University Uro-gynecological Department. All patients provided a detailed case history and underwent standard uro-gynaecological work-up. UI was assessed according to the International Continence Society (ICS) Criteria and graded

according to Ingelman-Sundberg. The clinical uro-gynaecological examination staged the POP according to the Halfway system and POP-Q test. Each patient underwent a standard urodynamic study (uroflowmetry, subtracted dual-channel filling cystometry with provocation test - cough and Valsalva's manoeuvre - and a pressure flow study), in accordance with ICS standards. VLPP, MUCP and a trans-rectal pelvic statics ultrasound were performed. All the patients were invited to fill out the self-administered IIQ. The answers were stratified according to age, type of pathology which patients were referred for, and educational status: low (none or primary school), medium (secondary school degree) and high (university degree). Statistical analysis was performed using the descriptive tests, the Kruskal-Wallis Test and the Spearman correlation test;  $p < 0.05$  was set as significant.

## **RESULTS**

Only 47.1% of patients answered all questions, 35.5% answered more than 5 questions and 13.3% did not answer more than 5 questions; 41.6% of patients belonging to low educational status answered all the questions against the 47.6% and 58.5% of patients with medium and high educational status respectively; this difference was statistically significant ( $p 0.038$ ). Women with a low educational status replied more often to questions on embarrassment and urine odour, they frequently did not answer the following questions: ability to do the shopping, go to the cinema or concerts, travel for more than 30 minutes by car or on the bus, go on holiday, friendships, clothes, mood and sleep pattern; 45% of women with a medium level of schooling did not reply to questions about housework, health conditions, urine odour or embarrassment. Finally, women with a high educational status (university degree) did not answer question on sexual relationships, physical activity, ability to do the shopping, hobbies and pastimes, odour and embarrassment. Age and pathology patients were referred for did not influence the answer rate ( $p 0.123$ ). No significant correlation was found between POP and/or UI severity and answer score ( $p 0.10$ )

## **CONCLUSION**

One of the main problems with self-administered tests is that some questions are inevitably left unanswered and our findings seem to confirm that IIQ is not an exception to this general observation. The high number of unanswered questions, particularly by women with a low educational standard, was probably due to poor or non understanding of some parts of the test. This may not be surprising as the IIQ was developed with a reference population of highly educated women over 45 years of age.



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| <b>PERINEAL ULTRASOUND ASSESSMENT OF VESICO-URETHRAL MOBILITY BEFORE AND AFTER TVT-O</b>   |
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### INTRODUCTION AND AIM OF STUDY

Pelvic floor ultrasound is an investigational procedure able to document vesico-urethral morphological and functional changes at rest and during provocation tests in the management and surgical follow-up of female stress urinary incontinence (FSUI) and pelvic organ prolapse (POP).

Aim of this study is to assess these changes before and after transobturator (TVT-O) sling procedure.

### MATERIALS AND METHODS

Seventeen consecutive patients underwent TVT-O procedure for FSUI due to urethral hyper-mobility. An ultrasound examination was performed preoperatively and 3 months after the surgical procedure. The perineal ultrasound was performed with a 3.5- MHz curved array probe with a bladder filling of 300 ml of sterile saline. During perineal ultrasound the following urethral angles and rectangular coordinate system are employed: **x-axis** (the central line of the pubic symphysis); **y-axis** (perpendicular to the x-axis at the lower border of the symphysis); **Dx** (distance between bladder neck and y-axis); **Dy** (distance between bladder neck and x-axis); **Urethral angle ( $\alpha$ )** (formed by the axis perpendicular to a reference line, such as the central line of the symphysis, and by the urethral axis); **Retrovesical angle ( $\beta$ )**; **Pubourethral angle ( $\gamma$ )** (between the bladder neck and the inferior border of the symphysis and the central line of the same).

### RESULTS

The table below shows the comparison between ultrasound measures before and after TVT-O.

**Table**

| <b>Ultrasound parameter</b>            | <b>Before TVT-O<br/>Median<br/>(interquartile range)</b> | <b>After TVT-O<br/>Median<br/>(interquartile range)</b> | <b>P value</b> |
|--|--|---|----------------|
| <i>Dx at rest</i>                      | 9 (6.25-11,8) mm   | 10 (8.5-15.5) mm  | NS             |
| <i>Dx on Valsalva</i>                  | 16.5 (13.25-19.5) mm                                     | 17 (13-17.5) mm   | NS             |
| <i>Dy at rest</i>                      | 16 (15-18.5) mm  | 14 (12.5-21) mm   | NS             |
| <i>Dy on Valsalva</i>                  | 4.5 (0-5.75) mm  | 5 (3-12) mm   | NS             |
| <i><math>\alpha</math> at rest</i>     | 15° (12°-19°)  | 18° (13.5°-23°)   | NS             |
| <i><math>\alpha</math> on Valsalva</i> | 53° (37.5°-61.75°)                                       | 39° (25.5°- 54°)  | NS             |
| <i><math>\beta</math> at rest</i>      | 145° (141°-146.5°)                                       | 140° (136°-148°)  | NS             |
| <i><math>\beta</math> on Valsalva</i>  | 147° (136.5°-158.75°)                                    | 151° (147.5°-157.5°)                                    | NS             |
| <i><math>\gamma</math> at rest</i>     | 126° (107°-130.5°)                                       | 132° (115.5°-144°)                                      | NS             |
| <i><math>\gamma</math> on Valsalva</i> | 170° (159.25°-179°)                                      | 155° (143.5°-176.5°)                                    | 0.04           |

**DISCUSSION AND CONCLUSION**

Pelvic floor ultrasound may be considered as a simple, mini-invasive and useful tool capable to assess urethral mobility in female patients after FSUI surgery. Our data suggested that TVT-O tape would not seem to negatively affect the bladder neck and the proximal urethra mobility. In contrast, the tape might slightly decrease middle urethra mobility during Valsalva manoeuvre.

## **CORRELATION BETWEEN VOIDING LOWER URINARY TRACT SYMPTOMS AND FLOW-PRESSURE ANALYSIS IN OLD-OLD PATIENTS**

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### **INTRODUCTION AND AIM OF THE STUDY**

The role of urodynamics in assessing and managing lower urinary tract dysfunctions (LUTD) in the old-old population (aged  $\geq 75$  years) is still debated. The disparity in the assessment of LUTD between lower urinary tract symptoms (LUTS) and urodynamic diagnosis is particularly evident with voiding symptoms (1). Aim of the study was to determine whether LUTS correlate with urodynamic flow-pressure observations in patients aged  $\geq 75$  years, using ICS definitions (2).

### **MATERIALS AND METHODS**

The flow-pressure traces and reports of 59 consecutive patients (31 males, 28 females) aged  $\geq 75$  years were retrospectively evaluated. The mean age was 79.4 years (range 75-93). The indications for urodynamics are summarised in table 1.

The following LUTS were considered: hesitancy, poor and intermittent urinary stream, terminal dribbling, straining; furthermore, the presence of an indwelling catheter or intermittent catheterisation was taken into account, as well as the pelvic organ prolapse (POP) only for women.

The following flow-pressure parameters were evaluated: bladder outlet obstruction (BOO), detrusor underactivity (DU), detrusor acontractility (DA), abdominal straining and post-void residual (PVR). BOO and detrusor contractility were measured in men by means of the pressure-flow diagram according to Schäfer, while in the female group the Blaivas-Groutz nomogram was used in order to define the presence of urethral obstruction.

Data were statistically analysed with the chi-square test. Statistical significance was set at  $p < 0.05$ . All data analyses were performed using SPSS release 12.0 for Windows.

**Table 1. Indications for urodynamics**

| Indication                         | Male (%)  | Female (%) | Whole population (%) |
|------------------------------------|-----------|------------|----------------------|
| Urinary retention                  | 20 (64.5) | 2 (7.1)    | 22 (37.3)            |
| Mixed urinary incontinence         | 0         | 11 (39.2)  | 11 (18.6)            |
| Voiding symptoms                   | 5 (16.1)  | 5 (17.8)   | 10 (16.9)            |
| Stress urinary incontinence        | 5 (16.1)  | 3 (10.7)   | 8 (13.6)             |
| Urgency ± urinary incontinence     | 1 (3.2)   | 5 (17.8)   | 6 (10.2)             |
| Recurrent urinary tract infections | 0         | 2 (7.1)    | 2 (3.4)              |

**RESULTS**

Patients clinical characteristics and cystometric observations made during the voiding phase are shown in tables 2 and 3, respectively.

**Table 2. Patients' clinical characteristics according to the gender.**

| Clinical characteristics     | Male (%)  | Female (%) | Whole population (%) |
|------------------------------|-----------|------------|----------------------|
| Hesitancy                    | 5 (20.5)  | 8 (36)     | 13 (29.7)            |
| Poor stream                  | 9 (34.6)  | 16 (49.1)  | 25 (43.2)            |
| Intermittent stream          | 5 (25.6)  | 15 (48.2)  | 25 (39.1)            |
| Straining                    | 8 (29.5)  | 13 (43.9)  | 21 (38)              |
| Terminal dribbling           | 7 (39.7)  | 14 (47.4)  | 21 (44.3)            |
| Indwelling catheter          | 18 (33.3) | 0          | 18 (13.5)            |
| Intermittent catheterization | 1 (2.6)   | 2 (4.4)    | 3 (3.6)              |
| POP                          | 0         | 12 (42.8)  | 12 (20.3)            |

**Table 3. Flow-pressure observations according to the gender.**

| Flow-pressure observations | Male (%)  | Female (%) | Whole population (%) |
|----------------------------|-----------|------------|----------------------|
| BOO                        | 12 (38.7) | 1 (3.6)    | 13 (22)              |
| DU                         | 17 (54.8) | 8 (28.6)   | 25 (42.3)            |
| DA                         | 10 (32.2) | 7 (25)     | 17 (28.8)            |
| Straining                  | 17 (54.8) | 17 (60.7)  | 34 (57.6)            |
| PVR ≥ 100 ml               | 20 (64.5) | 5 (17.8)   | 25 (42.3)            |

In the whole group a statistically significant correlation was found between the following variables: hesitancy with PVR  $\geq$  100 ml ( $p=0.03$ ); poor stream with DU/DA ( $p=0.014$ ); intermittent stream with DU/DA ( $p=0.020$ ); terminal dribbling with PVR  $\geq$  100 ml ( $p=0.03$ ).

As far as it concerns the difference between genders, BOO, DU and PVR  $\geq$  100 ml were significantly more common in males ( $p=0.002$ ,  $0.03$  and  $0.002$ , respectively). In the female group the only case of urethral obstruction was seen in a woman with POP. On the contrary, poor stream, intermittent stream and terminal dribbling were significantly predominant in females ( $p=0.03$ ,  $0.002$  and  $0.03$ , respectively).

## **DISCUSSION AND CONCLUSION**

The present study showed that in the whole group of old patients voiding LUTS, such as slow and intermittent stream, are significantly correlated with impaired detrusor contractility, while LUTS were not significantly correlated with BOO, that was confirmed to be rare in the women. BOO, DU/DA, and incomplete bladder emptying predominantly affected the male group. On the contrary, slow stream, intermittency and terminal dribbling were significantly more frequent in women than in men.

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**RELATIONSHIP BETWEEN PELVIC FLOOR MUSCLE DURABILITY AND ABDOMINAL MUSCLE ACTIVITY CHANGING ANKLE INCLINATION IN STANDING POSITION: PRELIMINARY RESULTS FROM A PILOT STUDY ON CONTINENT AND INCONTINENT WOMEN**

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**INTRODUCTION AND AIM OF THE STUDY**

A standing posture including various ankle positions might effectively facilitate pelvic floor muscle (PFM) activity through enhanced pelvic tilt in women with stress urinary incontinence (SUI), and an ankle dorsiflexion at 15° could be the best position to increase PFM activity (PFMa) although it is very uncomfortable. No data are available on healthy female population and on abdominal muscle (AM) activity. We carried out this study to assess the durability of aspect in women with and without FSUI.

**MATERIALS AND METHODS**

A total of 66 women were enrolled: 31 (mean age 40 years, range 28-49) complained of FSUI (Group A), and 35 (mean age 26, range 18-35) were healthy volunteers (Group B). Exclusion criteria were: musculoskeletal problems; previous major abdominal or pelvic surgery; severe diseases; diabetes mellitus, a body mass index >30 kg/m<sup>2</sup>; intrauterine device implantation; pelvic organ prolapse, menopause. An electromyographic (EMG) biofeedback instrument using surface electrodes was employed to measure changes in PFM and abdominal muscle (AM) activity.

During EMG recordings, each subject was asked to perform PFM 5s-contractions while assuming the following different upright positions: horizontal standing (HS), ankle dorsiflexion standing at 5° (5D), 10° (10D) and 15° (15D), and ankle plantar flexion standing at 5° (5P), 10° (10P) and 15° (15P). An adjustable basculant platform was used to passively set the ankle in each position. We analysed the following parameters: PFM activity at rest (rPFMa), during maximal contraction (mPFMa) and PFM durability (PFMd); AM activity at rest (rAMa), during maximal contraction (mAMa) and AM durability (AMd).

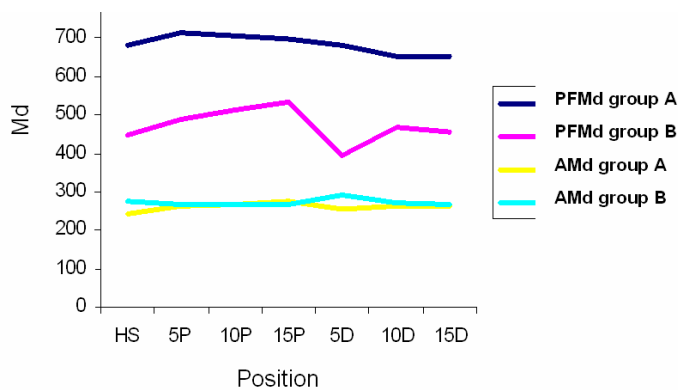
## RESULTS

The table below shows the mean values and the standard deviations of rPFMa, mPFMa and PFMd in the 7 analysed positions according to the two groups.

| Position       | rPFMa<br>(DS) $\mu$ V | mPFMa<br>(DS) $\mu$ V | PFMd(DS)<br>$\mu$ Vs |
|----------------|-----------------------|-----------------------|----------------------|
| <b>Group A</b> |                       |                       |                      |
| HS             | <b>19 (10)*</b>       | <b>344 (209)*</b>     | <b>682 (318)*</b>    |
| 5P             | 20 (10)               | <b>391 (200)*</b>     | <b>712 (332)*</b>    |
| 10P            | 21 (13)               | <b>374 (187)*</b>     | <b>706 (313)*</b>    |
| 15P            | 22 (12)               | <b>359 (192)*</b>     | <b>698 (343)*</b>    |
| 5D             | 25 (13)               | <b>359 (160)*</b>     | <b>680 (278)*</b>    |
| 10D            | 25 (10)               | <b>339 (170)*</b>     | <b>654 (283)*</b>    |
| 15D            | 28 (10)               | <b>338 (189)*</b>     | <b>653 (34)*</b>     |
| <b>Group B</b> |                       |                       |                      |
| HS             | <b>26 (16)*</b>       | <b>227 (117)*</b>     | <b>447 (224)*</b>    |
| 5P             | 23 (12)               | <b>265 (173)*</b>     | <b>489(323)*</b>     |
| 10P            | 23 (11)               | <b>265 (180)*</b>     | <b>511 (335)*</b>    |
| 15P            | 24 (12)               | <b>268 (170)*</b>     | <b>534 (331)*</b>    |
| 5D             | 27 (9)                | <b>215 (113)*</b>     | <b>392 (183)*</b>    |
| 10D            | 29 (11)               | <b>241 (146)*</b>     | <b>468 (256)*</b>    |
| 15D            | 31 (12)               | <b>232 (130)*</b>     | <b>454 (221)*</b>    |

**\*Significant differences between Group A and B**

The picture below shows the trend of PFMd and AMd according to the different analysed positions. No difference in terms of durability was found changing ankle inclination.





## **DISCUSSION AND CONCLUSION**

In order to achieve a more correct abdomino-perineal contraction, a variation of ankle inclination might have a role in the improvement of abdominal muscle activation. These results might change in incontinent women at the end of a PFM training period, after learning how to activate the correct abdomino-perineal synergy.

**THE EFFECT OF ANKLE INCLINATION IN SUPINE POSITION ON THE ABDOMINAL AND PELVIC FLOOR MUSCLES DURABILITY IN WOMEN WITH AND WITHOUT STRESS URINARY INCONTINENCE: PRELIMINARY RESULTS FROM A PILOT STUDY**

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**INTRODUCTION AND AIM OF THE STUDY**

Pelvic floor muscle (PFM) training is the first line treatment for female stress urinary incontinence (FSUI). Actually, it is not well known which the best position to facilitate PFM activity is. No data are available on the effect of ankle inclination on PFMa in the supine position according to abdominal influences. We carried out this study to assess this aspect in women with and without FSUI.

**MATERIALS AND METHODS**

A total of 66 women were enrolled: 31 (mean age 40 years, range 28-49) complained of FSUI (Group A), and 35 (mean age 26, range 18-35) were healthy volunteers (Group B). Exclusion criteria were: musculoskeletal problems; previous major abdominal or pelvic surgery; severe diseases; diabetes mellitus, a body mass index >30 kg/m<sup>2</sup>; intrauterine device implantation; pelvic organ prolapse, menopause. An electromyographic (EMG) biofeedback instrument using surface electrodes was employed to measure changes in PFM and abdominal muscle (AM) activity. During EMG recordings, each subject was asked to perform PFM 5s-contractions while assuming the following different supine positions:

ankles relaxed in a neutral position (NP), hips and knees at right angle (GF),

ankles dorsiflexed at 0°(0sD), 5° (5sD), 10° (10sD), and 15° (15sD), and ankles plantar flexed at 5°(5sP), 10°(10sP) and 15° (15sP). We analysed the following parameters: PFM activity at rest (rPFMa), during maximal contraction (mPFMa) and PFM durability (PFMd); AM activity at rest (rAMa), during maximal contraction (mAMa) and AM durability (AMd).

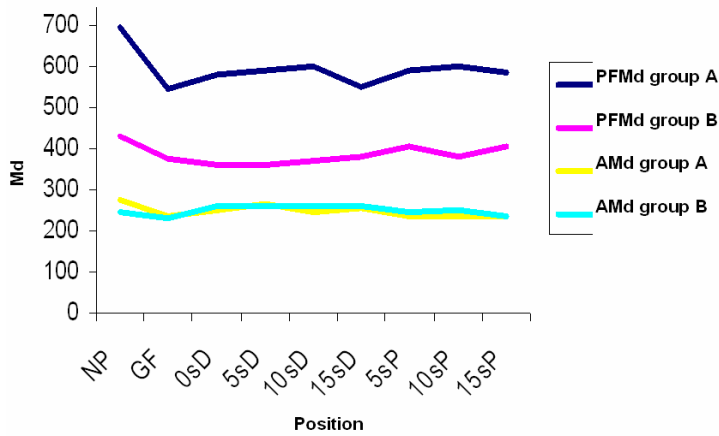
## RESULTS

The table below shows the mean values and the standard deviations of rPFMa, mPFMa and PFMd in the 9 analysed positions according to the two groups.

| Position       | rPFMa<br>(DS) $\mu$ V | mPFMa<br>(DS) $\mu$ V | PFMd (DS)<br>$\mu$ Vs |
|----------------|-----------------------|-----------------------|-----------------------|
| <b>Group A</b> |                       |                       |                       |
| NP             | 14 (13)               | <b>373 (201)*</b>     | <b>697 (332)*</b>     |
| GF             | 16 (9)                | <b>241 (122)*</b>     | <b>543 (227)*</b>     |
| 0sD            | 17 (11)               | <b>290 (148)*</b>     | <b>578 (272)*</b>     |
| 5sD            | <b>14 (10)*</b>       | <b>310 (173)*</b>     | <b>588 (307)*</b>     |
| 10sD           | 17 (15)               | <b>319 (175)*</b>     | <b>598 (306)*</b>     |
| 15sD           | 16 (13)               | <b>271 (151)*</b>     | <b>548 (265)*</b>     |
| 5sP            | 14 (13)               | <b>290 (172)*</b>     | <b>589 (318)*</b>     |
| 10sP           | 12 (12)               | <b>305 (171)*</b>     | <b>601 (299)*</b>     |
| 15sP           | 13 (13)               | <b>293 (147)*</b>     | <b>583 (245)*</b>     |
| <b>Group B</b> |                       |                       |                       |
| NP             | 13 (6)                | <b>225 (139)*</b>     | <b>432 (242)*</b>     |
| GF             | 18 (9)                | <b>184 (74)*</b>      | <b>376 (138)*</b>     |
| 0sD            | 18 (8)                | <b>184 (91)*</b>      | <b>361 (189)*</b>     |
| 5sD            | <b>20 (12)*</b>       | <b>193 (107)*</b>     | <b>358 (181)*</b>     |
| 10sD           | 16 (6)                | <b>196 (108)*</b>     | <b>372 (197)*</b>     |

**\*Significant differences between Group A and B**

The picture below shows the trend of PFMd and AMd according to the different analysed positions. No difference in terms of durability was found changing ankle inclination.



## DISCUSSION AND CONCLUSION

In the supine position an ankle dorsiflexion at 0° may improve PFM tone, whilst ankles relaxed in a neutral position may facilitate a better maximal PFMa. Moreover, in incontinent women a lack in the abdomino-perineal coordination has been observed.