Methods Seven healthy volunteers, one male and six female (five with normal vaginal deliveries and one nulliparous female), were studied to measure changes in vaginal and anal pressure and electromyographic activity (EMG) during a series of abdominal manoeuvres. Recordings were made with subjects in supine lying using vaginal and anal pressure probes (Incare, Hollister, USA), tollowed by EMG recordings with a vaginal probe (Periform, Neen, UK) and anal adhesive surface electrodes (Medtronic, Denmark). In the male subject anal pressure and EMG were measured simultaneously. Abdominal activity was monitored by a pair of surface electrodes placed on the lateral abdominal wall, and by ultrasound recording of abdominal muscle changes. Recordings of gastric pressure with the transducer inserted via the nose, were made in one female and the male subject. Hip adductor and gluteal muscle activity were also monitored in these two subjects, using pairs of surface electrodes. Subjects performed two repetitions of 1) maximal PF contraction 2) gentle abdominal isometric muscle contraction 3) moderate abdominal isometric contraction 4) strong abdominal isometric contraction. Further EMG recordings were made in standing. These were1) maximal PF contraction 2) abdominal wall relaxation and gentle contraction in an upright position 3) abdominal wall relaxation and gentle contraction in a forward lean position. EMG data was expressed as an increase in amplitude from baseline as a percentage of that recorded during a maximal voluntary PF contraction. EMG and pressure data were compared between conditions using a repeated measures one-way analysis of variance (ANOVA) and Duncan's multiple range test. Results EMG of the PF muscles, recorded from both anal and vaginal electrodes, increased above resting level with all of the abdominal contractions. Analysis of the hip adductor and gluteal muscle EMG indicated that the PF EMG was not affected by cross-talk from these muscles. The increase in PF EMG with a strong abdominal contraction was significantly greater than with the gentle and moderate contractions. The EMG recorded with the vaginal electrode during the strong abdominal contraction was no different from that recorded during a maximal PF muscle contraction. Vaginal and anal pressures increased in a similar manner to the EMG changes. Gastric pressure increases commenced after the onset of anal and vaginal pressures. Both in upright and forward lean standing positions vaginal EMG showed a decrease in activity from baseline when the abdomen was relaxed and an increase with a gentle abdominal hold. Conclusions The specific abdominal exercises used in this study result in PF muscle activity in healthy subjects. The onset of PF EMG prior to the onset of gastric pressure indicates that the response of these muscles must be preprogrammed by

of PF EMG prior to the onset of gastric pressure indicates that the response of these muscles must be preprogrammed by the central nervous system to contribute to intra-abdominal pressure and maintain continence. The results indicate that abdominal exercises such as these could be used to maintain PF muscle support, but further research is required to confirm the successful application of such exercises for rehabilitation in symptomatic subjects.

<u>References</u> 1. Activation of the abdominal muscles is a normal response to contraction of the pelvic floor muscles. ICS abstract 117, Yokohama. 1997.

2. Activation of pubococcygeus during a variety of isometric abdominal exercises. ICS abstract 115, Yokohama 1997.

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PARAURETHRAL CONNECTIVE TISSUE STATUS OF POSTMENOPAUSAL WOMEN WITH GENITAL PROLAPSE WITH AND WITHOUT GENUINE STRESS INCONTINENCE

An effective closure of the female urethra in stress situations is dependent on an integrated action of various anatomical intra- and extraurethral structures. The most important extraurethral structures - from a functional aspect - are the suburethral vaginal wall, the pubourethral ligaments, the pubococcygeus muscles and the paraurethral connective tissue. An important ingredient in the supportive structures of the genitourinary region is fibrous connective tissue, consisting mainly of collagen and structural glycoproteins. Hence, defects in the actual connective tissue – in particular the paraurethral connective tissue that connects the aforementioned structures to each other and to the urethra – will bring about an ineffective urethral closure. Female genuine stress incontinence (GSI) may be caused by defective connective tissue per se and / or by a disconnection to extraurethral structures, whereby the urethra cannot be closed at stress situations. Genital prolapse may or may not be associated with GSI, or may even

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be masked due to the extent of pelvic floor relaxation. However, little knowledge exists why some patients with prolapse develop GSI and some do not.

Aims of study: The status of paraurethral connective tissue in postmenopausal female patients was investigated for relevant changes of paraurethral connective tissue between continent and incontinent women with genital prolapse.

Methods: Before pelvic reconstructive surgery patients underwent a complete urogynecologic assessment including history, urinalysis, evaluation of residual bladder volume, medium fill cystometry, urethral pressure profile measurements (UPP) at rest and during stress, and a clinical stress test in the supine and standing position. A pronounced prolapse was repositioned during UPP measurements and during a clinical stress test to assess or exclude GSI. All patients underwent pelvic floor reconstructive surgery (anterior colporrhaphy: n=19, sacrospinous

fixation: n=8). Patients with proven GSI received additional tension-free vaginal tape procedure.

During pelvic floor reconstructive surgery biopsies from both paraurethral regions were obtained. Biopsies were investigated for localization and distribution of both collagen (types I, III, IV, V, VI) and glycoproteins (fibronectin, laminin, vitronectin) using immunofluorescent microscopic techniques. Frozen biopsies were incubated with primary polyclonal antibodies (1:25 dilution in PBS) at room temperature for 60 minutes. Three washings with a 0.2% PBS-BSA solution and exposure to Trimethylrhodamin-isothiocyanat (TRITC)-conjugated second antibodies (swine antirabbit immunglobulins [Dakopatts, Denmark] diluted 1:20 dilution in PBS) were performed to visualize the protein. After another washing with PBS-BSA solution, photographs of the samples were taken (Kodak Ektachrome 400) for documentation.

Results: GSI was present in 10/19 women (Group A, mean age: 56,2 years), whereas 9/19 patients (Group B, mean age: 59,6 years, p > 0.05) were continent.

Irrespective of the presence or absence of GSI all types of collagen (I,III, IV, V, VI) were found in the biopsies of the whole study group. The tissues of Group A patients showed a marked weaker immunohistochemical reaction of type I, III and VI collagen compared with the specimen of group B patients.

No difference of Type IV and V collagen was observed between the biopsies of group A and B patients. Type V collagen was located in the subepithelial connective tissue zone of the stroma, touching the basement membrane and forming a fibrillar meshwork. Type IV collagen was selectively found in the zone of basal membrane and vessel walls

Among the structural glycoproteins fibronectin and laminin were found in the specimen of all patients. However staining of fibronectin was less pronounced than that of collagen. Nevertheless, fibronectin was distinctly found in the extracellular matrix. The stroma revealed a fine fibrillar matrix reaching to the basement membrane. Laminin showed a similar distribution in the basement membrane as type IV collagen.

Vitronectin was be observed in the paraurethral connective tissue of all group B patients, whereas this glycoprotein was lacking in the biopsies of group A women. In vitronectin positive tissues the stained structures were granular-like or fibrous and sometimes amorphous. A striking association of vitronectin with elastic fibres was seen.

Conclusions: There is a complex architecture of the extracellular matrix in the female paraurethral region with marked differences between postmenopausal, continent women and patients with GSI, irrespective of the presence of pelvic floor relaxation. Our findings suggest a selective and altered metabolism of connective tissue in the paraurethral region responsible for the onset of GSI in patients with pelvic floor relaxation.

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Title (type in CAPITAL LETTERS, leave one blank line before the text):
ALTERATIONS IN THE STRUCTURAL AND MOLECULAR CHARACTERISTICS OF THE
PELVIC FLOOR TISSUES IN DIABETES MELLITUS (DM).

Introduction and objective- The absence of insulin and insulin like growth factors (IGF) has been reported