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Biomechanical Properties of the Pelvic Floor and its Relation to Pelvic Floor Disorders

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Abstract

Pelvic organ prolapse and stress urinary incontinence remain a clinical challenge as they have unclear pathophysiology and suboptimal treatments. These common pelvic floor disorders (PFD) are characterized by the weakening of the pelvic floor supportive tissues that are directly related to their biomechanical properties. Characterizing the biomechanical properties of the pelvic floor tissues has been the focus of recent studies and researchers are using tools that are not always well understood by clinicians. Therefore, the aim of this review is to provide an overview of the most used methods to test the passive biomechanical properties of the human pelvic floor tissues. We also summarize recent findings from studies looking into the passive properties of the pelvic floor in pelvic floor disorders using the ex vivo tensile test and emerging in vivo techniques. Together, these studies provide valuable quantitative information about the different biomechanical properties of the supportive tissues of the pelvic floor under normal and pathological conditions. Results from ex vivo tests provide valuable data that needs to be correlated to the in vivo data and the clinical manifestations of the symptoms of the PFD. As more research is conducted we will obtain an enhanced understanding of the effect of age, PFD, and treatments on the biomechanical properties of the pelvic floor. This information can contribute to better identify individuals at risk, improve clinical diagnosis, and develop new treatments to advance clinical practice.

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1. Introduction

Pelvic organ prolapse (POP) and stress urinary incontinence (SUI) are public health concerns and important pelvic floor disorders (PFD) affecting millions of women worldwide. Both are caused by the weakening of pelvic floor (PF) supportive tissues and occur independently or coexist. They remain a challenge because they are multifactorial conditions with unclear pathophysiology and suboptimal treatments. A woman's lifetime risk for either POP or SUI surgery is as high as 20% [1], with a substantial reoperation rate due to recurrence [2].

The supportive soft tissues within the PF are a combination of muscles, fascias, and ligaments working together to keep the pelvic organs in place in a highly dynamic environment. Biomechanical tools are used to understand how tissues function together to provide support and resist deformations. The biomechanical properties of a tissue can be active or passive and can be measured using well characterized ex vivo destructive techniques or new emerging in vivo methods. Most studies looking into the biomechanical properties of the human PF supportive tissues have reported the passive biomechanical properties. Such properties allow tissues to transmit loads or resist deformations without generating external forces. These properties are important because the PF supportive tissues are loaded and deformed by different activities and conditions that can be physiological (eg, age, parity, walking, jumping, breathing), or pathological (eg, vomiting, obesity, previous surgery, chronic coughing). Thus, changes in the biomechanical properties of these tissues and loading environment may help to better understand PFD. These fundamental understandings contribute to identify individuals at risk and to improve clinical practice and diagnosis resulting in new treatments.

There are many different terminologies and means to report the material properties of tissues. As studies grow, researchers are using different tools to characterize the biomechanical behavior of tissues that can sometimes be confusing. Therefore, the aims of this review are: (1) to provide an overview of the most frequently used methods to test the biomechanical properties of PF soft tissues in humans, (2) to review the current literature on the passive mechanical properties of the different support tissues of the human female PF, and (3) to put this information in perspective of PFD.

2. Why should we study tissue biomechanics in the pelvic floor?

The PF tissues are a combination of muscles, fascias, and ligaments that form a hammock-like support at the bottom of the abdomino-pelvic cavity that are attached to the pelvic bones. They have two basic functions: (1) to provide support to the pelvic organs (ie, the bladder, vagina, uterus, and rectum), and (2) to facilitate intercourse, vaginal delivery, storage of stool, and voluntary defecation and urination. Successful PF support is able to resist the loading environment without resulting in a pathology (eg, SUI, POP, fecal incontinence, etc.). Therefore, PF support is dependent on the loads these PF tissues experience and the biomechanical properties of the tissues themselves. A natural variation in the loading environment or material properties may make certain individuals more at risk for developing a PFD than others. For example, we can examine two individuals with high loads on their PF tissues. The first individual may not develop PFD because their tissues are able to resist these mechanical loads (forces), while the second individual has weaker tissues and thus progresses to develop a PFD. Therefore, it becomes critical to accurately understand the loading environment that these PF tissues are exposed to, and to directly characterize the biomechanical properties of the PF tissues. With this knowledge, physicians could adapt treatments according to the individual's needs for a better long-term patient care.

2.1. The loading environment of the PF

The PF is constantly being loaded by intra-abdominal pressure (IAP) because of its anatomical location and daily activities. The IAP is a physiological load that is transmitted from the lungs and diaphragm through the abdominal cavity and eventually onto these PF tissues. This load can fluctuate with passive or active compression of the abdominal wall, breathing, load bearing, coughing, laughing, etc. Higher loads would mean higher force increments in the IAP and therefore changes in the mechanical loadings to the PF.

Changes in the IAP have been recorded in the bladder and are assumed to be a good representation of the pressures received by the PF. The maximum intrabladder pressure for nonpregnant healthy volunteers has been reported to be as high as 347 cmH_2O during coughing and vomiting in the right lateral decubitus position [3]. Straining a stool typically leads to peak pressures of 100 cmH_2O over several seconds [4]. Obesity can further increase the baseline of the IAP by 19 cmH_2O [5]. These conditions (eg, chronic coughing, constipation, and obesity) are related to well identified risk factors of PFD and represent a profound load on pelvic organs. In addition, POP and SUI have been strongly linked with injury incurred during parturition [6]. Unsurprisingly, the maximum pressures exerted on the PF muscles are during the second stage of labor, where the already high basal levels due to the pregnancy itself, can be increased a further 194 cmH_2O [4]. Such pressures become higher than the increased IAP for coughing and straining present in nonpregnant women and may last for as long as 1 hr. The combination of these elevated loads, duration, and deformation presents a high risk for injury to the PF. Therefore, over the last decade the concept of biomechanics has become a focal point in understanding POP and SUI.

3. How are the biomechanical properties of the PF tissues being tested?

Understanding tissues' biomechanical properties facilitates access to tissues' function. Unlike traditional materials, biological tissues are complex in organization, function, and

behavior. Biomechanical properties contain both active and passive components. Active properties relate to tissue's ability to produce a force through muscle contractions, while passive properties represent a tissue's ability to resist deformation or transmit loads. Many structures contribute to the active and passive biomechanical properties to support the PF. Important contributors to the active properties are the levator ani muscle group and the vaginal wall that has a considerable amount of smooth muscle. There are some studies that have focused on the active properties of vaginal tissues in animal models, and in human samples [7–12]; however, here we will concentrate our discussion on the passive properties of human samples because these properties are more widely reported in the literature [13–24] and human samples are more clinically relevant. Passive properties allow tissues to transmit loads or resist deformations without generating external forces and are divided into *structural* and *mechanical*. *Structural* properties include loads and displacements (ie, elongation) that are directly measured during an experiment, while *mechanical* properties refer to computed properties such as stresses and strains. The *structural* and *mechanical* properties can be investigated with a traditional uniaxial tensile test (Fig. 1). The following sections will focus on understanding how to utilize biomechanics to characterize the passive biomechanical properties of the PF with emphasis on the uniaxial tensile test.

3.1. *Structural properties*

Typically, uniaxial tensile tests are performed in a destructive manner, meaning that the tissue is tested until failure/rupture. The recorded measurements from the load-cell and cross-head are used to generate a load-displacement curve (Fig. 1B), which is used to determine a specimen's *structural* properties. For biological tissues the load-displacement curve can be broken into a nonlinear or *toe-region*, followed by a linear region, and then a yield and failure region. In urogynecology, the toe-region is frequently referred to as the comfort zone (an indication on how tissues react to small displacements, ie, physiological daily activities), and the linear region is referred to as the stress zone (an indication on how tissues react to exceptional stretching that are thought to be outside the normal range) [25]. The *structural* properties include specimen stiffness, which is the slope of the load-displacement curve. It can be calculated in the toe-region (comfort zone stiffness) and linear region (stress zone stiffness). Following the linear region there is typically a yield region, where the tissue begins to fail until the entire tissue ruptures. At rupture, the

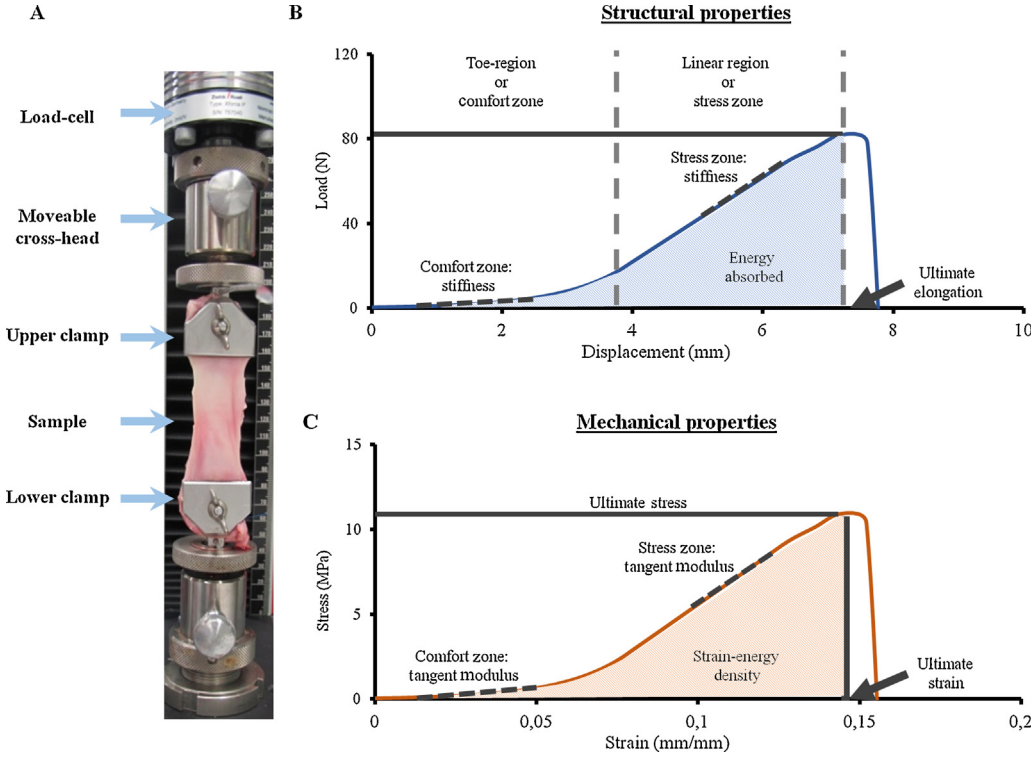

Fig. 1 – Representative structural and mechanical properties of a specimen from a uniaxial tensile test. Structural properties allow us to determine the biomechanical properties of a tissue or complex of tissues, while mechanical properties allow us to gain insight into the quality of a tissue by considering the size and geometry of the tissue. (A) An example of a tissue sample clamped on either side of a traditional uniaxial tensile test. Typically, one clamp remains fixed while the other clamp, in-line with a load-cell, is attached to a moveable cross-head. After clamping, the tissue is commonly dissected to a mid-substance length to width ratio of 5 or greater to create a uniform stress and strain environment at the midsubstance [52,53]. As the cross-head moves the specimen is loaded and a load-cell records load and the displacement. (B) Illustrates the different regions of the load-displacement curve. The toe-region, or comfort zone, illustrates initial non-linear response of the tissue to a uniaxial load. Highlighted are the structural properties: comfort zone and stress zone stiffness (N/mm), ultimate load (N), and corresponding ultimate elongation (mm), and the energy absorbed (N*mm). (C) Stress–strain curve from a uniaxial tensile test. Parallel to structural properties, mechanical properties include a comfort and stress zone modulus (MPa), the ultimate stress (MPa), ultimate strain, and strain-energy density (MPa).

ultimate load and elongation are reported. Lastly, energy absorbed can be calculated, defined as the area of the load-displacement curve until failure/rupture of the specimen.

3.2. Mechanical properties

While *structural* properties are valuable and frequently the best available means to characterize a group of tissues, they do not inform us about the *mechanical* properties. The *mechanical* properties are obtained from a stress–strain curve and can be derived from the same uniaxial test. Stress (σ) is defined as a load per unit of the cross-sectional area (CSA) of the specimen (Eq. (1)).

$$\sigma = \frac{F}{CSA} \quad (1)$$

For a simple uniaxial test, strain (ε) is the change in length ($\Delta L = L - L_o$) divided by the initial length (L_o) of the specimen (Eq. (2)).

$$\varepsilon = \frac{\Delta L}{L_o} \quad (2)$$

It should be noted that when the specimen has a sufficient aspect ratio (length/width), the mechanical properties should be calculated from the center of the specimen, where a uniform distribution of stress and strain occurs [26–28]. These *mechanical* properties are related to the accuracy of calculating stress and strain, and there are multiple methods that can be used to determine the CSA and strain which have been described by others [29–32].

The specimen's *mechanical* properties are derived from the stress–strain curve (Fig. 1C), and are analogous to the *structural* properties (Fig. 1B). If a specimen displays a simple linear response it would be defined as linear-elastic material and the Young's modulus, a measure of tissue stiffness, would be the slope of the curve. However, most biological tissues have stress–strain curves with a time-dependent behavior because they are viscoelastic materials, so a tangent modulus can be calculated both in the comfort and in the stress zones. The tangent modulus is defined as the slope of a stress–strain curve, and can be taken at any given range, thus it is important to clearly define how and where a tangent modulus was calculated. We can also calculate the ultimate stress, ultimate strain, and the strain-energy density.

3.3. Difference between structural and mechanical properties

A main difference between the *mechanical* and *structural* properties is that *mechanical* properties normalize to individual specimens' dimensions. This is important because *structural* properties between two specimens may appear similar but their *mechanical* properties can be very different. This often happens in healing tissues when more collagen is produced to overcome an injury but the type and/or organization of collagen is different [28,31]. It is also possible that multiple samples have different *structural* properties, but in fact have similar *mechanical* properties. Figure 2 shows an example of two linear-elastic tissues with the same *mechanical* properties but different *structural* properties due to geometric variations. In this example, the slope of the line is called the Young's modulus, which is reserved for describing linear-elastic materials that do not have a toe-region. Therefore, the behavior of a typical biological tissue (Fig. 1B and C) and a linear-elastic material (Fig. 2) are remarkably different, which is why the term tangent modulus is frequently used to describe the slope of biological tissues. Here, we illustrate how these changes in size could easily occur due to specimen preparation (ie, differences in clamping length or dissection), and should be taken into account when comparing *structural* properties between specimens and across studies. This simplified case used linear-elastic materials; however, this effect is common in biological testing when not carefully controlled in the experimental design (eg, the stiffness values of materials are different, but the tangent modulus values are the same) and could be a critical issue when using the *structural* mechanical properties of the tissues to develop new meshes or implants to treat POP.

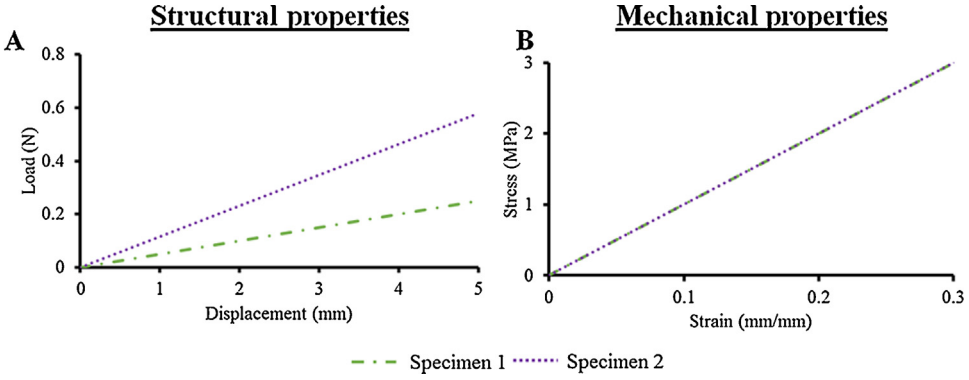

Fig. 2 – Theoretical linear-elastic specimens with different (A) structural properties but (B) the same mechanical properties. (A) Represents the load-displacement curve of these two specimens that have different lengths and cross-sectional areas (CSA): specimen 1 has a length of 20 mm and a CSA of 0.1 mm^2, while specimen 2 has a length of 19 mm and CSA of 0.22 mm^2 (this difference in CSA represents a 0.1 mm difference in each the width and thickness measurement). From the load-displacement graph, specimen 1 and specimen 2 have a stiffness of 0.05 N/mm and 0.12 N/mm, respectively. However, when we calculate the Young's Modulus this approximate 2× difference in stiffness equates to the same Young's modulus (10 MPa). (B) Thus, when normalized to their respective CSA and length, these specimens have a similar stress–strain relationship.

3.4. Hyperelasticity

The above examples are similar in a sense as each uses a linear relationship to describe the behavior of the tissue (Young's modulus) or quantify a part of the stress–strain response (tangent modulus). However, it is clear from Figure 1 that most tissues are nonlinear; therefore, it is important to examine alternative approaches to characterize a tissue's stress–strain relationship. To model complex behaviors of biological tissues, researchers often use the concept of hyperelasticity. This approach allows to mathematically relate the stress–strain relationship of a material through a strain energy density function and has been covered in detail in various biomechanical publications [33,34].

One of the earliest hyperelastic models developed to describe rubber materials was the Mooney-Rivlin constitutive relationship. Similar to the material properties described above, a Mooney-Rivlin material is assumed to be isotropic, elastic, and incompressible; however, this model captures the nonlinearity of biological tissues and can describe the entire stress–strain relationship. Rubod et al [15], introduced a Mooney-Rivlin like model to describe the behavior of pelvic floor tissues. In this relationship, stress (σ) is defined as a function of two parameters (C_0 and C_1) and tissue stretch (λ).

$$\sigma = \left(\lambda - \frac{1}{\lambda^2}\right) * \left(C_0 + 2 * C_1 * \left[\lambda^2 + \frac{2}{\lambda} - 3\right]\right) \quad (4)$$

Above, we described how to calculate stress (Eq. (1)) and strain (Eq. (2)) from a uniaxial test. We can directly calculate tissue stretch from strain ($\boldsymbol{\lambda} = \varepsilon + 1$), thus we only have two unknown parameters in our stress–strain relationship C_0 and C_1. It is then possible to use a fitting function from various programs (ie, Matlab, Mathematica, or SAS) to determine the values for C_0 and C_1 that best describe the stress–strain curve. There are various methods of acquiring these parameters using these programs [35].

Due to this model's simplicity (ie, only requiring two parameters to describe the stress–strain relationship), each of the parameters in this constitutive model has an easy interpretation: C_0 is interpreted as the stiffness of a tissue in the low strain or deformation range; and C_1 indicates the stiffness of the tissue at the higher strain regions. To illustrate how C_0 and C_1 influence the biomechanical response we again created an idealized sample (Fig. 3). Thus, these parameters give us a base of how to compare tissues throughout the nonlinear stress–strain relationship.

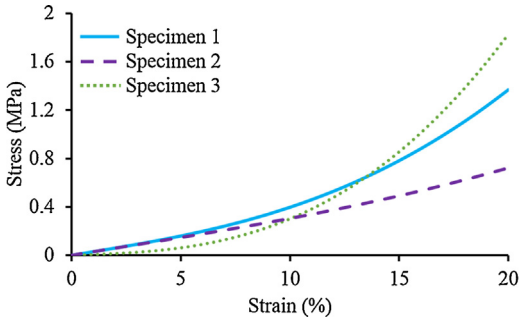

Fig. 3 – Idealized example to illustrate how to interpret the role of C_0 and C_1 coefficients when applying a modified Mooney-Rivlin relationship to describe the biomechanical response of a specimen to uniaxial tension. Here, we are looking at a stress–strain curve of three theoretical samples with various C_0 and C_1 coefficients. In this example, specimen 1: C_0 = 0.5 MPa and C_1 = 4 MPa, specimen 2: C_0 = 0.5 MPa and C_1 = 1 MPa, and specimen 3: C_0 = 0.1 MPa and C_1 = 8 MPa. We can see that the same C_0 (specimen 1 and specimen 2) results in the same stress–strain relationship in the low strain region (< 5% strain). However, specimen 3, which has a lower C_0, is less stiff in this region. Examining the influence C_1, we see that a larger C_1 leads to a more nonlinear stress–strain response as a tissue is stretched under uniaxial tension. A C_1 of zero would results in a completely linear response. We can also note that compared with specimen 1, specimen 3 has a lower stiffer initially due to a lower C_0; however, as these theoretical tissues are stretched Specimen 3 becomes more stiff a due to a higher C_1.

4. Passive biomechanical properties of the human PF in PF disorders

4.1. Ex vivo techniques

The most used test is the uniaxial tensile test, but studies have shown high variability between the values reported to characterize the biomechanical properties of the tissues (Table 1). Even though such variations could be related to physiological differences among women, it is clear that the lack of standardized research protocols is also an issue. In particular, harvesting the testing tissue is a very critical step because mechanical properties are sensitive to the location and size of a sample. Due to the rarity of the sample, researchers usually take as much of a sample as possible to maximize the utility of the tissue, which can introduce data errors. Therefore, comparing data between studies from different groups is difficult. However, because of its importance, we have endeavored to understand the biomechanical properties of the PF, not only to find the best material for the repair, but also to avoid more tissue damage over the long term.

Ruboud et al [36], reported the effect of different sample preparations in sheep vaginal tissues when performing uniaxial tensile tests in PF tissues. This initial work formed the bases of other research comparing the biomechanical properties of vaginal tissues [17], pelvic ligaments [24], and the effect of age on different tissues of the PF [23].

Different pelvic tissues within the same woman have different degrees of stiffness and experience large levels of strain before rupturing. Under uniaxial tension, tissues experienced strain up to 20% for the vagina, 30% for the rectum, and 80% for the bladder [19]. Uterosacral ligaments and broad ligaments were more rigid and less extensible than the vaginal tissues in elderly [20]. In contrast, the mechanical behavior between the ligaments and the vagina in young women were not statistically different [23]. It seems that in young women the uterosacral ligaments and the vagina would react similarly to small displacements (every day movements) and to exceptional stretching (ie, birth trauma or chronic coughing) because they have a similar degree of stiffness at low and high strain levels. With age the biomechanical properties of the ligaments and vaginal tissues become more rigid and there is a mismatch

Table 1 – Studies reporting passive biomechanical properties of pelvic floor supportive tissues using the ex-vivo tensile test technique

Reference	Tissue	*N*	Group, *n*	Sample source	Mean age (yr)	Reported measurement	Main findings
Goh et al, 2002 [13]	Vagina	18	POP-post, 10 POP-pre, 8	POP patients	69 41	Elastic modulus Deformation	Postmenopausal tissues ↑elastic modulus than premenopausal.
Lei et al, 2007 [14]	Vagina	20 23	POP-post, 12 POP-pre, 9	POP patients	61 46	Elastic modulus (MPa) Poisson's ratio Maximum elongation Maximum fracture	Elastic modulus: POP > non-POP; ↑ severe type and moderate type > mild type. Maximum elongation and maximum fracture: non-POP > POP; mild type > severe type and moderate. Connective POP tissues are less elastic and stiffer. Biomechanical properties: POP < control group.
Rubod et al, 2008 [15]	Vagina	10	POP, 5 Non-POP, 5	POP patients Female cadavers	74 76	Strain at rupture (%) Stress at rupture (MPa)	Nonlinear elastic behaviour with large deformation = hyperelasticity.
Jean-Charles et al, 2010 [16]	Vagina anterior and posterior	40	POP, 30 Non-POP, 10	POP patients Female cadavers	66 76	Rigidity (MPa)	POP > non-POP.
Gabriel et al, 2011 [17]	Aponeurosis, skin, and vagina	11	Non-POP, 11	Female cadavers	79	Rigidity (MPa) Ultimate strain at rupture (%)	Aponeurosis > vagina > skin. Aponeurosis > skin > vagina.
Martins et al, 2011 [18]	Bladder	13	Non-POP, 13	Female cadavers	45	Stiffness (MPa) Maximum stress (MPa)	Bladder young stiffer than old.
Rubod et al, 2012 [19]	Vagina, bladder, and rectum	5	Non-POP, 5	female cadavers	75	Rigidity (MPa) Ultimate strain level (%)	High deformations: vagina > tissue > bladder. Vagina anterior < posterior. Vagina < rectum < bladder. Anisotropic behavior of the bladder.
Rivaux et al, 2013 [20]	Broad, round, and uterosacral ligaments	13	Non-POP, 13	Female cadavers	N.M.	Rigidity (MPa)	Uterosacral ligaments > round. Ligaments > broad ligaments.
Martins et al, 2013 [21]	Round and uterosacral ligaments	15	Non-POP, 15	Female cadavers	45	Stiffness (MPa) Maximum stress (MPa)	Stiffness and maximum stress: uterosacral > round ligaments. Nulliparous had uterosacral ligaments with lower stiffness and maximum stress.
Martins et al, 2013 [22]	Vagina	55	POP, 40 Non-POP, 15	POP patients Female cadavers	64 45	Stiffness (MPa) Maximum stress (MPa)	POP anterior > posterior wall. non-POP anterior < posterior wall. POP > non-POP.
Chantereau et al, 2014 [23]	Vagina, bladder, rectum, uterosacral ligament, round ligament, and broad ligament (Group 1) Vagina, bladder, and rectum (Group 2) Uterosacral ligament, round ligament, and broad ligament (Group 3)	6	Group 1, 6 Group 2, 5 Group 3, 13	Female cadavers	29 75 84	Rigidity (MPa)	Bladder < vagina and ligaments. Vagina old > young. Uterosacral and round ligaments old > young. Low deformations: bladder and rectum old > young.
Lopez et al, 2015 [24]	Vagina	28	BMI < 25, 13 BMI > 25, 15	POP-post patients	63 64	Maximal tangent modulus (MPa) Yield strain (%) Yield stress (MPa) Strain energy at tissue specimen's yielding point (MPa) Failure strain (%) Failure stress (MPa) FSE (MPa)	BMI > 25: shorter toe region for the stress–strain responses and ↑stiffness in biomechanical properties of POP tissues. BMI > 25: ↑tangent moduli, ↑yield strain, ↑yield stress, ↑failure stress and ↑FSE. Women had vaginal hormone cream twice a wk 2–3 wk before operation.

BMI = body mass index; FSE = strain energy at tissue specimen's failure point; MPa = megapascal; N.M = not mentioned; POP = pelvic organ prolapse; post = postmenopausal; pre = premenopausal.

between them: they will react differently to daily physiological loads. Since the ligaments are more rigid and less extensible than the vaginal tissues, the loads that are transmitted in the vaginal tissues in young women and older women are different. If they are too high, the vaginal wall can even be injured, which can make this tissue even more vulnerable, complicated by the fact that there is impaired wound healing in elderly people [37,38]. Such mismatches in tissue biomechanics might partially explain the increase in POP occurrence in elderly women.

Others reported that age and/or parity affect the biomechanical properties of other supportive tissues from the PF. Higher stiffness and maximum stress of the uterosacral ligaments compared to the round ligaments were reported in parous women compared to nulliparous women [21]. However, no association between the biomechanical properties of ligaments and age, body mass index, and menopausal status was found, which could be related to the sample size (Table 1).

The stiffness reported in these studies can be comparable to the reported rigidity at large strain levels (C_1), as they are derived from similar stress–strain experimental curves from uniaxial tensile tests. In this sense, these results are consistent with observations by Rivaux et al [20], as they also found that the uterosacral ligaments were stiffer than the round ligaments. In addition, Chantereau et al [23], reported an increased "rigidity" with age in the uterosacral and round ligaments, which was not confirmed by Martins et al [21].

Aside from parity, biomechanical properties can vary spatially based on anatomical location. In the vagina, differences between the anterior and posterior vaginal wall have been reported. In vaginal tissues from women younger than 50 yr of age, the posterior seems to be stiffer than the anterior wall [22]. Others have seen the opposite in vaginal tissues from specimens from women older than 50 yr, where the anterior seems to be stiffer than the posterior wall [16,19,22]. One study also examined women with or without POP, and reported higher stiffness in the anterior compared to the posterior vaginal wall of women with POP [22].

Although it is difficult to exclude that the increased stiffness found in tissues from women with POP is part of a physiological aging process, there is evidence indicating a correlation between severity of POP and the increased stiffness in the anterior vaginal wall [14,22], even when using different biomechanical tests [39]. It seems that the changes in the biomechanical properties of the anterior vaginal wall due to age and parity, might make this site more vulnerable which is consistent with clinical data that shows that the most frequent kind of POP occurs in the anterior compartment (cystocele) [40–42].

Biomechanical properties of supportive tissues from the PF provide valuable data; however, the ex vivo measurements require harvesting of relatively large tissue samples from patients. Moreover, PF supportive tissues from women without PFD are scarce as they are obtained from fresh cadavers or more typically from older patients, without clinical information. Therefore, new in vivo approaches might provide a good alternative with the extra advantage that they could be used in clinical practice.

4.2. *In vivo approaches*

To follow the changes and progress into pathology overtime would be advantageous in the development of enhanced in vivo approaches to measure biomechanical properties of PF tissues. In vivo approaches are starting to emerge using pressure transducers, imaging methods or a combination of both (Table 2).

In 2007, Epstein et al [43], used dermatology suction-based devices to compare systemic and vaginal biomechanical properties of women with normal vaginal support and POP. Local but not systemic alterations were found in the biomechanical skin properties of women with POP compared to controls. Women with POP showed significantly more extensible vaginal mucosa than women with normal pelvic support.

In a follow-up study using a different probe, improvements in the in vivo biomechanical properties of the vaginal sidewall were shown in women undergoing sacral colpopexy [44]. Chuong et al [45], recently compared the prolapsed anterior vaginal wall with the suprapubic region (control site within the same woman) of postmenopausal women with cystocele using a similar suction device-like instrument. Results showed that the POP-site, in response to mechanical disturbance, had a lower capacity in elastic recoil, and a higher degree of energy loss compared to control tissues. This suggests that the POP tissues are more compliant than controls, have higher viscous damping and are less able to store recoverable energy upon distension [45].

The higher compliance found in POP tissues obtained with a suction device-like instrument [45] suggest that they are less stiff than controls, which are opposite to the results from ex vivo tests [14,22,39]. It is challenging to directly compare these results as these in vivo and ex vivo measurements do not assess the same material response properties of the vagina. For example, several of these ex vivo biomechanical tests examined the stiffness of the vagina while being stretched under cyclic loading. Whereas with the suction-like devices, other viscoelastic properties, such as tissue elasticity (the capacity of skin to regain its original form after a deforming force is removed) and tissue extensibility (the capacity of skin to be stretched), can be measured by recording the vertical change of the tissue while exposed to a constant mild vacuum in the device probe [43]. It is important to understand how ex vivo testing (where we have more control and freedom to examine various loading conditions), relates to different in vivo measurements before they can be used clinically.

To better understand the different forces sustained by the vaginal wall, a mixture between imaging and pressure sensors could be a better approach. The vaginal tactile imaging device was designed to be an evaluation method which also allows the assessment of elastic properties of the vaginal walls and imaging of differential pressures in real time [46]. Using the vaginal tactile imaging, researchers

Table 2 – Studies reporting passive biomechanical properties of pelvic floor supportive tissues using the in vivo techniques

Reference	Tissue tested	*N*	Group, *n*	Device	Mean age (yr)	Reported measurement	Main findings
Epstein et al, 2007 [43]	Vaginal skin, systemic skin	48	POP, 25 Non-POP, 23	DermaLab skin probe (both) Cutometer MPA 580 (vaginal skin)	56.5 56.9	Stiffness index Vaccum pressures: high-gate pressure and low-gate pressure	Vaginal mucosa: POP < non-POP. No differences in the systemic skin.
Epstein et al, 2008 [44]	Vaginal side wall	35	POP, 35 Before sacral colpopexy After sacral colpopexy	DermalLab USB skin probe	56.3	Viscoelasticity (MPa) Elasticity (MPa) Stiffness index (mbar) Extensibility time (s) Retraction time (ms)	Preoperative vs. postoperative: ↓viscoelasticity, ↓elasticity, ↓extensibility time and ↑retraction time.
Chuong et al, 2014 [45]	Anterior vaginal wall	22	Post-menopausal POP, 22 POP site (bladder neck) Non-POP site (suprapubic region)	BTC-2000TM cutometer-like instrument	N.M.	Peak uplift (u_{peak})[a] Residual uplift ($u_{residual}$) Rate of tissue recoil or tissue recovery rates (E/η) SE (stored) SE (recovered) Hysteresis or SE (loss) = SE (stored) – SE (recovered)	POP vs. non-POP sites: ↑ u_{peak}, ↑ $u_{residual}$, ↓ recovery rates, ↑ SE (stored), ↓ SE (recovered), ↑ SE (loss).
Egorov et al, 2010 [46]	Vaginal walls	13	POP, 7 Operated POP, 3 Non-POP, 3	VTI	N.M.	Rigidity Elasticity index (kg/deg) 3D imaging of tissue elasticity by real time visualization of pressure patterns.	↓ elasticity: posterior vs. anterior vaginal wall non-POP, and POP vs non-POP. ↑ rigidity in sites of implanted meshes.
Egorov et al, 2012 [47]	Vaginal walls	31	POP, 13 Non-POP, 18	VTI	60	Tissue elasticity (kPa). 3D imaging of tissue elasticity by real time visualization of pressure patterns.	↓ elasticity: POP vs non-POP.
van Raalte et al, 2015 [48]	Vaginal walls	20	POP-Q I, 4 POP-Q II, 7 POP-Q III, 4 POP-Q IV, 1 Non-POP, 4	VTI	55.5	Pressure Pressure gradient Dynamic pressure during muscle contraction	The reported measurements were correlated to pelvic floor conditions and night be used for further characterization.
Smith et al, 2013 [49]	UST	17	POP, 17	Own tripod-mounted computer –controlled linear servoactuator to quantify the force-displacement behavior of the uterine cervix.	54.5	Ligament "stiffness" on different cervix locations per patient = Δ traction force/Δ cervical displacement between minimal and maximal force.	POP-Q point C was strongly correlated with cervix location at rest and at maximum traction force, only 19% of the variation in POP-Q point C location was explained by ligament stiffness. The cervix location in the OR at minimal traction lay below POP-Q point C value in three-fourths of the women.
Luo et al, 2014 [50]	UST	14	POP, 14	Same as Smith et al, 2013 [49]	53.9	Stiffness (N/mm) Energy absorbed (J) Normalized final force = relaxation force/peak force	UST has hyperelastic behavior and can stretch to twice the maximum force of 17.8N.

Table 2 (*Continued*)

Reference	Tissue tested	*N*	Group, *n*	Device	Mean age (yr)	Reported measurement	Main findings
Swenson et al. 2017 [51]	UST, apical/vaginal support	52	Normal/normal, 14 Normal/prolapse, 11 Prolapse/prolapse, 27	Same as Smith et al, 2013 [49]	47.1 52.9 59	Cervix location under different tractions forces Apical support stiffness (N/mm) Cervix displacement (mm)	With traction force from 1–18N 50% of women in normal/POP group and 59% of women in the group POP/POP group had abnormal apical support. Apical support stiffness from 1–18N was lowest in women from the prolapse/prolapse group.

Δ = delta; 3D = three dimensional; NM = not mentioned; POP = pelvic organ prolapse; POP-Q = POP-Quantification; SE = effective strain energy density stored; UST = uterine suspensory tissue; VTI = vaginal tactile imaging.
[a] Authors used a Voigt model.

have reported decreased elasticity in the vaginal walls from women with POP [46,47]. In a follow up study, 11 parameters were identified as potential markers to characterize POP including pressure, pressure gradient, and dynamic pressure response during muscle contraction [48]. These findings are interesting and future iterations of this device may prove to be a useful diagnostic tool, but in order to be clinically relevant, this study must be properly validated.

In 2013, Smith et al [49], reported the development of a device to measure the stiffness of the uterine suspensory tissues in vivo, by applying a continuous force while simultaneously recording the cervical displacement. Results showed that the uterine suspensory tissues exhibited visco-hyperelastic behavior and it could stretch to twice their initial length under the maximum force applied of 17.8N [50], which is in line with reported hyperelastic behavior of the ligaments from tensile tests [20,21]. Recently, Swenson et al [51] used the same device to try and objectively help to make a decision about a hysterectomy by finding relationships between normal apical support, vaginal support, cervix location, and apical support stiffness.

Although it is still early to provide the specific parameters which would support a clinical decision making, their data showed a substantial discrepancy between cervix locations during apical support testing suggesting that some properties of cervical support are not sufficiently captured by the POP-quantification examination [51]. However, it is important to note that the experimental device is used under anesthesia while the POP-quantification examination is performed when the patient is awake, which might explain the discrepancy found in the results. The relevance of this discrepancy is unclear.

The mentioned in vivo measurements are important and have the potential to be very useful as clinical tools for diagnostics and for evaluation of treatments. However, it is important to realize that they are relatively new tools and often require making priori assumptions about a tissue's behavior that can affect the outcome parameters. Therefore, it is still important to characterize individual tissues using well established ex vivo tests that provide accurate data to be compared to in vivo tests to check for consistency, correlations, or agreement between these testing modalities. To make such comparisons we need mathematical models that take into account different components of the pelvic floor supportive tissues and can predict their mechanical behavior as a whole because ex vivo tests are limited to small isolated tissue biopsies and in vivo tests assess tissues complexes in situ.

In summary, the most widely used method to test the passive biomechanical properties of human pelvic floor support tissues is the uniaxial tensile test, a well-characterized ex vivo technique. Such tests are providing important data showing that age and POP make both ligaments and vaginal wall tissues stiffer compared to controls. However, ex vivo techniques require the extraction of tissues and studies are limited by the sample size, site, and lack of controls. New emerging in vivo approaches have the potential to be very useful clinical tools for diagnostics and for evaluation of treatments for PFD. Nevertheless, they still need to be

validated in large female cohorts, and data needs to be correlated to data from well-established ex vivo tests with the aid of mathematical models.

5. Conclusions

PFD are common conditions characterized by the weakening of the PF supportive tissues. Identifying the changes in the biomechanical properties of the support tissues will help to understand PFD. The basic concepts from the most frequently used methods to test the passive biomechanical properties of the different support tissues help to understand the pathomechanism of the PFD. The results from initial ex vivo tests correlated to the emerging in vivo data and the clinical symptoms enhance the understanding and provide us with important insights about the effects of age, PFD, biomechanical properties of the PF tissues, and related treatments. With the gathered data of the biomechanical properties, individual risks can be identified, clinical diagnoses improved, and new treatments developed which collectively advance clinical practice.

Conflicts of interest

The authors have nothing to disclose.

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