Urol Res (1982) 10:285–291

Urological Research

Experimental Study on Bladder Wall's Strain in Vesical Function

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Accepted: March 30, 1982

Summary. The literature is reviewed and the applicability of Laplace's law to the urinary bladder considered as a sphere is critically discussed. Experiments were performed on live rabbits, on isolated bladders and on rubber balloons to study vesical adaptation and response to filling. From the data obtained, the authors propose "strain", rather than tension, as a characteristic quantity of bladder function and indicate the possible application of these experimental results clinically.

Key words: Urinary bladder, Micturition, Cystometrogram, Urodynamics.

Introduction

Since the first investigations on the physiology of the urinary bladder, attempts to apply units of measure have been made.

Mosso and Pellacani [18] first determined the pressure/volume P(V) characteristic during the vesical filling phase and interpreted the resulting diagrams in terms of tone, i. e. as a function of the bladder wall's properties; they also called the phenomenon of negligible increases in pressure related to measurable increases in the bladder's volume "adaptation"[1]. Sherrington [23] compared vesical pressure response with the postural reflex of skeletal muscles, assuming that an interaction with spinal centres was able to activate a reflex phenomenon which allowed the bladder to maintain almost constant pressure during decreases in volume.

Denny Brown and Robertson [10] noted a slight fall in intravesical pressure during rest periods following partial filling; they called this property "adaptation" and argued that it resulted from an interaction between an excitatory reflex (tone) and that of an inhibitory one (adaptation), always present even during the normal filling phase. The same adaptation pattern was described in patients with spinal cord injuries, so that the authors inferred that pressure variations were only related to the activity of the nervous plexuses of the bladder wall.

Langworthy et al. [17] ascribed the lowered vesical capacity in spinal animals to a lower "adaptability"[2] and assumed that bladder tone, as well as that of skeletal muscles, was under the control of the central nervous system.

Ruch and Tang [22], starting from the results obtained by Nesbit et al. [21] performed various investigations in healthy as well as in spinal animals finding a similar P(V) pattern; from their results they suggested that the CNS influenced only the micturition reflex and not the tone. They deduced that the pressure curve was merely due to the physical characteristics of the bladder wall and introduced a new quantity, tension, i. e. the strength exerted upon the bladder wall to contain a certain amount of fluid at a certain pressure level. To determine it, they applied Laplace's law, considering the bladder like a sphere:

$$T = \frac{P}{2}\sqrt[3]{V\frac{3}{4}}$$

approximated to: $T = 0.312 \quad P\sqrt[3]{V}$

However they did not consider the variations in the bladder wall's thickness during the filling phase, hence their data appear insufficient. Therefore, it is necessary to determine a unit of measure of bladder function which takes this variation into account.

[1] The term "adaptation" has been used with different meanings by different authors.

[2] This term was used to indicate an inhibitory reflex by which the bladder could bear an increase in fluid volume without a proportional increase in pressure.

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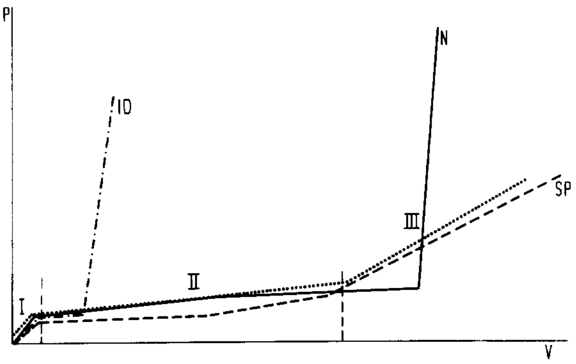

Fig. 1. CMG's pertaining to Fig. 1: *N* = normal subject; *ID* = intercollicular section; *SP* = spinal section

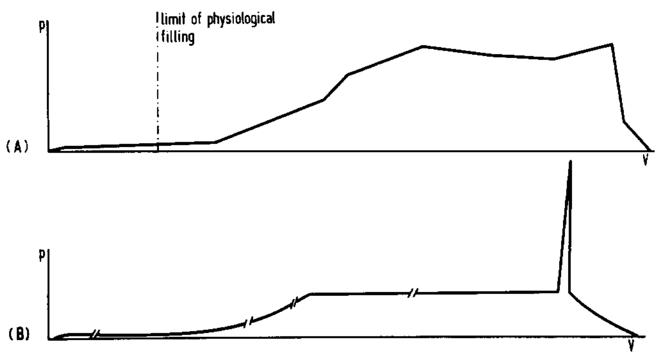

Fig. 2. P(V) characteristics in the bladder extracted from the rabbit (*A*) and in the rubber ballon (*B*)

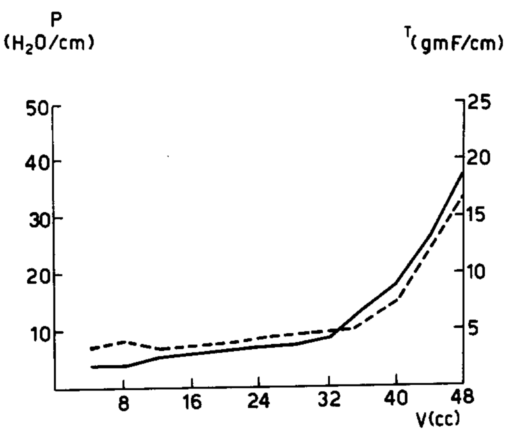

Fig. 3. The T(V) characteristic

Investigative Techniques for the Bladder

There are essentially four clinical and laboratory methods for studying the bladder: cystomanometrics, uroflowmetrics, cystography and cystoscopy. Figure 1 shows the P(V) characteristics to which we will refer in order to explain our experimental studies; these CMG's show that:

a) CMG typically consists of three parts:

1. A notable increase in pressure corresponding to an initial increase in volume (segment I), probably due to intra-abdominal pressure.

2. A nearly flat prolonged segment (segment II), related in some authors' opinion to the detrusor tone; normally it is followed by micturition.

3. In the absence of micturition segment III appears, probably representing passive distension of the bladder wall. In case of spinal sections, segment III can start at smaller or greater volumes compared with the micturition threshold.

b) No large variations in pressure are detectable in cases with spinal lesions or in decerebrated animals; the only difference is in the micturition threshold. In spinal animals, experiments show prolongation of segment II and a complete absence of segment III.

c) There is no linear correlation between volume and pressure. The slight increase in pressure associated with segment II is irrelevant compared with pressure variations exerted upon the bladder by abdominal viscera during postural movements; this fact probably indicates that the control on the bladder is not mediated by pressure.

d) The CMG's in Fig. 1 demonstrate that pathological situations only influence the micturition threshold and not pressure levels; therefore it is possible that vesical control during the filling phase is independent of brain or medullary centres, but a reflex phenomenon at lower levels cannot be excluded.

In order to try and answer these questions it is necessary to evaluate the following items:

1. Is the P(V) characteristic peculiar to the urinary bladder or common to other bodies?

2. Does the bladder present similar or different properties after it has been removed?

In order to clarify these aspects, CMG's have been performed on a rubber balloon filled with a saline solution and on a bladder just extracted from a rabbit after death.

Figure 2 shows the resulting diagrams: they are almost identical and similar to normal curves. This confirms the hypothesis that there is not a neurogenic control on the bladder filling pressure. Tension is the parameter that can be accepted to explain bladder function.

In fact certain smooth muscles, if submitted to tension, are able to contract autonomously. The T(V) characteristic, obtained by applying Laplace's law (Fig. 3), demonstrates that, with equal increases in volume, greater variations result compared with the P(V) characteristic. However, Laplace's law does not take into consideration the variations in thickness which can be considerable. In fact bladder muscles in a rabbit have an approximate volume of 3 cm^3; the filling volume varies from 20 ml to 44 ml; consequently bladder wall thickness undergoes a reduction of six times

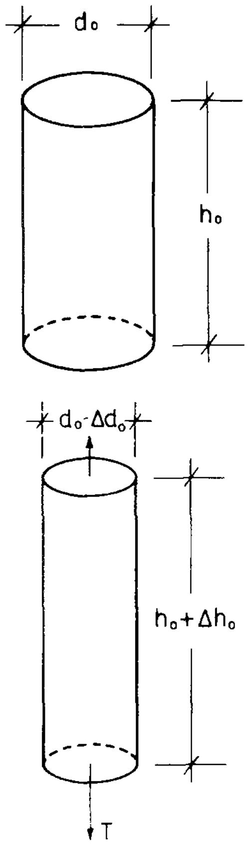

Fig. 4. Variations in the dimensions of a test-bar submitted to traction

from 3 mm to 0.5 mm. To neglect this consideration limits the application of Laplace's law to specific cases. Hence a new quantity has been proposed: strain (σ), i. e. the relation between tension and thickness of the wall on which tension is exerted.

How to Calculate σ

Assuming that the bladder is a membrane with a uniform thickness S, submitted to an internal pressure P, strains σ_1 and σ_2 on the wall are given by the following formulae:

$$\frac{\sigma_1}{R_1} + \frac{\sigma_2}{R_2} = \frac{P}{S} \tag{1}$$

$$\sigma_1 = \frac{PR_2}{2S} \tag{2}$$

where R_1 and R_2 are the radii in the direction respectively of strains σ_1 and σ_2.

Formula (1) is given by the equilibrium condition of a wall surface in the direction of its normal *n*.

Formula (2) is given by the equilibrium condition of a spherical bowl in the direction of its axis.

From (1) and (2) we have:

$$\sigma_2 = \frac{PR_2}{S}\left(1 - \frac{R_2}{2R_1}\right) \tag{3}$$

Now assimilating the bladder to a sphere:

$$R_1 = R_2 = R$$

hence (1), (2) and (3) become:

$$\frac{\sigma_1 + \sigma_2}{R} = \frac{P}{S} \tag{1'}$$

$$\sigma_1 = \frac{PR}{2S} \tag{2'}$$

$$\sigma_2 = \frac{PR}{2S} \tag{3'}$$

then:

$$\sigma_1 = \sigma_2 = \frac{PR}{2S} \tag{4}$$

How to Determine R and S

During the vesical filling phase both radius R and thickness S vary; these quantities are related to:

- the filling volume V_R
- the volume of the bladder wall's musculature V_M

It is known that material deformation of a body may occur with or without volume variations. To explain this phenomenon, consider Fig. 4: initially the volume of the test-bar is:

$$\bar{V}_0 = \frac{\pi d_0^2 h_0}{4}$$

If subsequently the test-bar is submitted to a traction strain (T), the following dilatations will result:

$$\epsilon_h = \frac{\Delta h_0}{h_0}$$

$$\epsilon_d = -\frac{1}{m}\epsilon_h = -\frac{\Delta d_0}{d_0}$$

and the dimensions will become:

$$h_1 = h_0 + \Delta h_0$$

$$d_1 = d_0 - \Delta d_0$$

and the volume will be:

$$\overline{V}_1 = \frac{\pi(d_0 - \Delta d_0)^2 (h_0 + \Delta h_0)}{4}$$

Now two possibilities are given:

1) $\overline{V}_1 = \overline{V}_0$

2) $\overline{V}_1 \neq \overline{V}_0$

In the first case the deformation occurs at a constant volume; in the second one it occurs at a variable volume. Then we can indicate:

$$\overline{V}_1 = \overline{V}_0 + \Delta\overline{V}_0$$

and the $\Delta\overline{V}_0/\overline{V}_0$ ratio, as a function of material and strain, is given by the relation:

$$\frac{\Delta\overline{V}_0}{\overline{V}_0} = \frac{3(m-2)}{2}\frac{\sigma_t}{E} \qquad (5)$$

where σ_t = strain; m = Poisson's coefficient; E = Young's modulus.

When Poisson's coefficient has a value m = 2, formula (5) becomes:

$$\frac{\Delta\overline{V}_0}{\overline{V}_0} = 0$$

and deformation can occur at an almost constant volume. Now, in a plastic range it is always m = 2, while in an elastic range the maximum value is m = 4. However, assuming that vesical dilatation occurs without volume variation, it has been considered m = 2; the same approximation is currently used also for elastic materials, considering the small variation range of m (2 ÷ 4).

In conclusion R and S have been determined by using the following formulae:

$$R_m = \frac{\sqrt[3]{\frac{3}{4\pi} V_R} + \sqrt[3]{\frac{3}{4\pi}(V_R + V_M)}}{2} \qquad (6)$$

$$S_m = \sqrt[3]{\frac{3}{4\pi}(V_R + V_M)} - \sqrt[3]{\frac{3}{4\pi} V_R} \qquad (7)$$

where R_m and S_m indicate the mean values.
By substituting (6) and (7) in (2′), we have:

$$\sigma_m = \frac{PR_m}{2S_m} = \frac{P}{4} \frac{\sqrt[3]{\frac{3}{4\pi} V_R} + \sqrt[3]{\frac{3}{4\pi}(V_R + V_M)}}{\sqrt[3]{\frac{3}{4\pi}(V_R + V_M)} - \sqrt[3]{\frac{3}{4\pi} V_R}} \qquad (8)$$

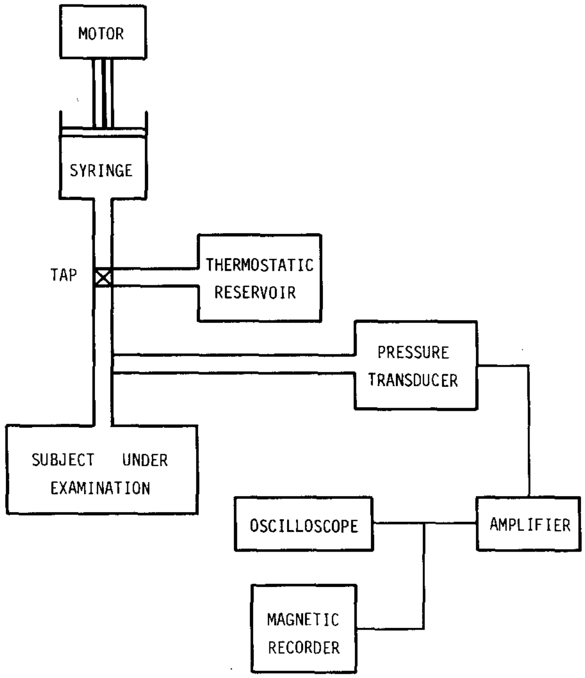

Fig. 5. Block diagram of the apparatus used for measuring the signal during experimentation

and simplifying:

$$\sigma_m = \frac{P(\sqrt[3]{V_R} + \sqrt[3]{V_R + V_M})}{4(\sqrt[3]{V_R + V_M} - \sqrt[3]{V_R})} \qquad (9)$$

The formula (9) has been used for the determination of strain (σ).

For $V_M \ll V_R$, equation (9) reduces to:

$$\sigma = 3\,PV_R(1 + V_M/6V_R)/2V_M.$$

Materials and Methods

The experimental investigations have been performed at the Institute of Veterinary Physiology of the University of Milan.

Only male rabbits were used in order to compare data obtained from male cats by other authors and because of the easier catheterisation of a male subject. In all the experiments (on live rabbits, on isolated bladders, on rubber balloons) a catheter was inserted and connected to a syringe and to a pressure transducer.

Pressure was detected by a Sanborn transducer (mod. 267 BC series 81) and the signal was amplified by a Sanborn amplifier (Carrier Preamplifier type mod. 350–1100 C) and visualised on a Tektronix 5031 oscilloscope then recorded on an FM magnetic recorder (mod. 3960 Hewlett & Packard) (Figs. 5 and 6). Filling was obtained with 2 ml of fluid (at a flow-rate of 165 ml/h) every minute, by a piston connected to a Braun Melbungen perfusor. The syringe was filled by a three-way system with a saline solution at 37 °C temperature in a thermostatic bath.

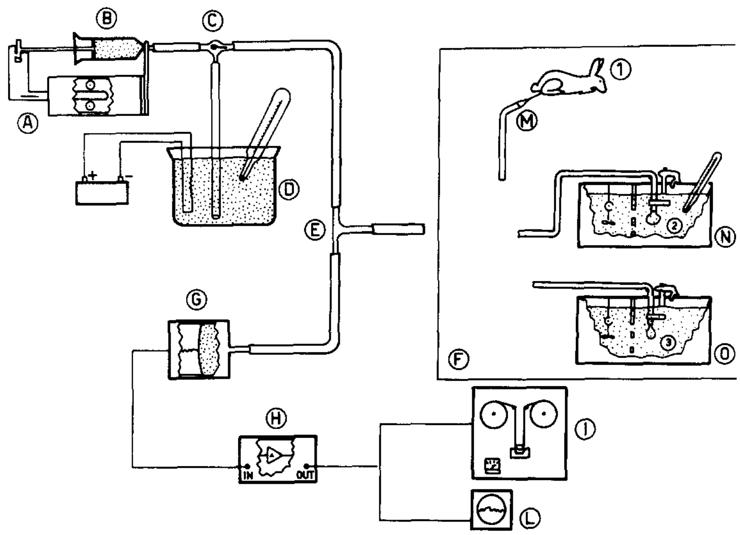

Fig. 6. Schema of the experimentation: A = perfursor; B = syringe; C = three-way tap; E = T connection F = subject under examination (*1.* rabbit; *2.* isolated bladder; *3.* rubber balloon); G = pressure transducer; H = amplifier; I = magnetic recorder; L = oscilloscope; M = catheter; N = thermostatic bath; O = ambient temperature bath

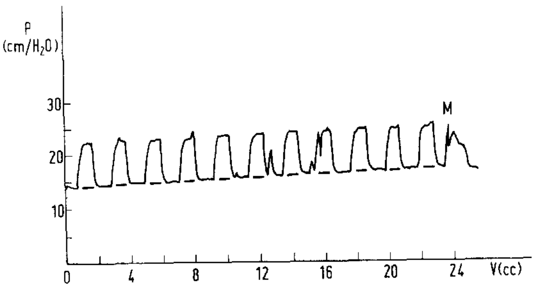

Fig. 7. A P(V) diagram during a test. —— = pressure variations as related to filling; - - - = "relaxed" pressure

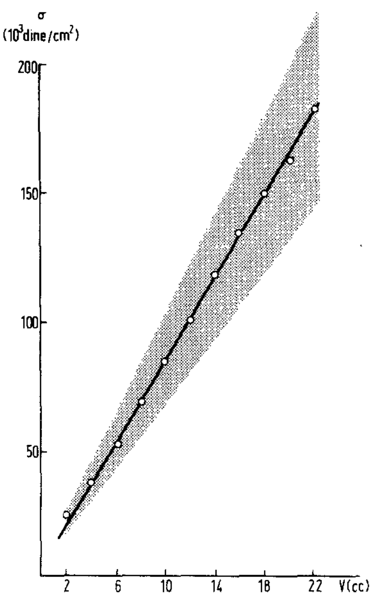

Fig. 8. Variation range of σ(V) characteristic

Tests on Rabbits

Eight male rabbits (New Zealand stock) with an average weight of 5 kg were used. They were not anaesthetised to avoid any interference on micturition reflex; 30 min before the test 0.1 mg/kg of Combelen (propionyl-promazine) was given IM to make the insertion of a Rusch catheter #6 F easier.

Immobilisation of the animals was obtained by using plaster bandages in order to prevent abrupt movements and consequential alterations of vesical pressure during the test; rectal temperature was 38 ± 0.2 °C during the whole experiment.

Tests on Isolated Bladders

After the previous experiments were performed, six of the eight rabbits were sacrificed by Nembutal then operated on to remove their bladders. The bladder, isolated from its nervous and vascular connections, was then divided from the urethra below the external sphincter and both ureters were ligated by #1 silk sutures. The wall volume was measured by putting the empty bladder into a measuring glass containing a known amount of saline solution.

The tests were performed keeping the bladder at 37 °C temperature in a thermostatic bath of oxygenated solution containing 95% O_2 and 5% CO_2.

Tests on Rubber Balloons

Rubber balloons placed in a bath containing a saline solution at ambient temperature were employed.

Experimental Results

In the P(V) diagram obtained by these experiments every peak coincides with a 2 ml filling (Fig. 7), except for the last one (M) which represents the start of micturition. Anomalous contractions of the subject tested are responsible for eventual distortions in the curve. The σ(V) characteristic has been obtained by using the formula (9). In the following diagrams, filling volumes in cc (ml) are indicated in abscissa, while strains in dyn/cm^2 (1 cm H_2O = 980.07 dyn/cm^2) are indicated in ordinate. Figure 8 shows the P(V) characteristic obtained from Fig. 7: it appears that strain increases in proportion to the filling volume, the curve being approximated to a linear regression:

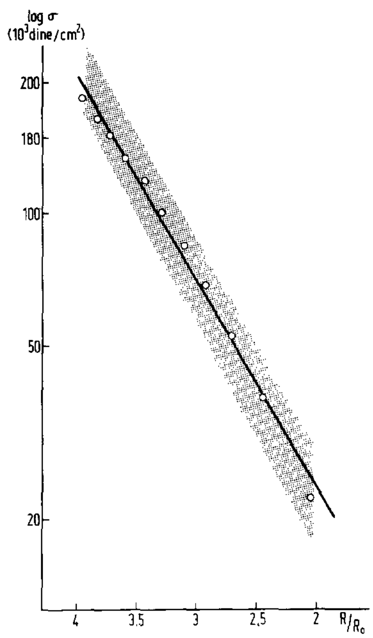

Fig. 9. Variation range of log $\sigma(R/R_0)$ characterstic

$\sigma = 8066.318V + 5228.727$

that gave a correlation index of $r^2 = 0.9995$.

Log σ (R/R_0) characteristic (Fig. 9) is reported in order to compare our data with that of other authors. This characteristic presents a decimal logarithmic scale in ordinate and the pure number R/R_0 in abscissa.

$$R_0 = \sqrt[3]{\frac{3V_M}{4\pi}}$$

This characteristic is also approximated by the regression:

$\log \sigma = 0.468\ R/R_0 + 3.442$

with a correlation index of $r^2 = 0.9992$.

Discussion

This work, compared with the hypothesis expressed by Ruch and Tang [22], confirmed that intravesical pressure variations during the filling phase are only related to the properties of the wall material. However, with regard to the quantity proposed as characteristic by these authors, it has been pointed out that they did not consider the variation that the bladder wall undergoes during the filling.

Therefore a new characteristic quantity has been proposed to give emphasis to this phenomenon: strain. In comparison with other work, it is significant that the log $\sigma(R/R_0)$ characteristic reported in Fig. 9 is in agreement with Alexander's data [1, 2].

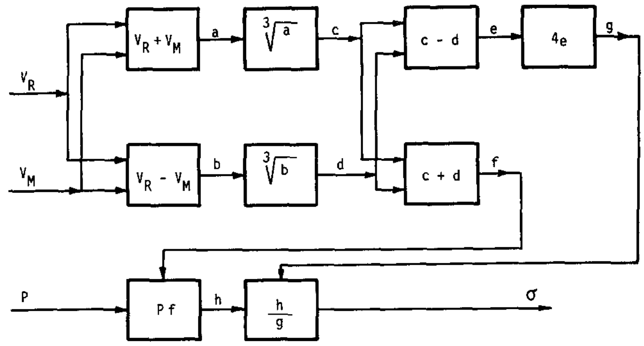

Fig. 10. Block diagram of the analogical circuit for the calculation of σ

In fact Alexander determined the strain-elongation ratio and the strength during relaxation phase using strips of mouse bladder, obtaining results similar to ours. Moreover, methods employed in this work do not require destruction of the organ tested and provide information from clinical tests.

With regard to the voiding phase, it has been noted in the clinical field that in patients with prostatic obstruction there is a notable decrease in the diameter but a relatively small decrease in flow (according to the Bernoulli's equation): it appears that the bladder musculature is submitted to very considerable strain. In fact this pathological feature is known as the "columnar bladder" of prostatic subjects. An analysis of "strain" can explain this alteration by observing that the structure of the bladder wall, submitted chronically to considerable strains, undergoes deterioration and loses its elastic properties. Recent studies on the vesico-urinary system [11–14, 24–26, 29, 30] have tended to apply electrical stimulation in the rehabilitation of micturition control. The analysis of strain can also offer a quantity significant for the state of the bladder to evaluate the opportunity for the use of a stimulator (still elastic or deteriorated tissue) as well as to determine the optimal intervals for stimulation.

Moreover these tests are useful to determine the micturiton threshold, which is also necessary for the use of anal or vaginal stimulators. The methods of analysis presented in this work are also applicable in the clinical field by using a cystomanometer.

The only difficulty is the elaboration of the resulting signal (pressure). On the other hand, this elaboration is possible by introducing into an analogue system not only the resulting signal (P), but also the filling volume (V_R) (function of a flow constant in time) and the bladder wall volume (V_M) (an initial constant determined by ultrasonic techniques).

The electronic apparatus used in this investigations consisted of a unit to elaborate strain; the resulting signal (σ) and the filling volume (V_R) were inserted respectively into the vertical (y) and into the horizontal (x) inputs of a writing machine (recorder on paper) (Figs. 10 and 11), thus obtaining a characteristic diagram.

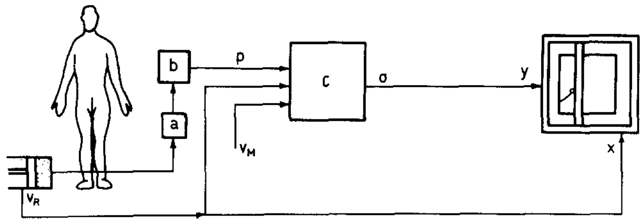

Fig. 11. Complete diagram for the calculation of strain using a CMG: *a* = pressure transducer; *b* = amplifier; *c* = analogical unit

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