Caveolae are involved in mechanotransduction during pulmonary hypertension

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Caveolae are involved in mechanotransduction during pulmonary hypertension. Am J Physiol Lung Cell Mol Physiol 310: L1078–L1087, 2016. First published March 25, 2016; doi:10.1152/ajplung.00198.2015.—Caveolae are stiff plasma membrane microdomains implicated in various cell response mechanisms like Ca2+ signaling and mechanotransduction. Pulmonary arterial smooth muscle cells (PASMC) transduce mechanical stimuli into Ca2+ increase via plasma membrane stretch-activated channels (SAC). This mechanotransduction process is modified in pulmonary hypertension (PH) during which stretch forces are increased by the increase in arterial blood pressure. We propose to investigate how caveolae are involved in the pathophysiology of PH and particularly in mechanotransduction. PASMC were freshly isolated from control rats (Ctrl rats) and rats suffering from PH induced by 3 wk of chronic hypoxia (CH rats). Using a caveolae disrupter (methyl-β-cyclodextrin), we showed that SAC activity measured by patch-clamp, stretch-induced Ca2+ increase measured with indo-1 probe and pulmonary arterial ring contraction to osmotic shock are enhanced in Ctrl rats when caveolae are disrupted. In CH rats, SAC activity, Ca2+, and contraction responses to stretch are all higher compared with Ctrl rats. However, in contrast to Ctrl rats, caveolae disruption in CH-PASMC, reduces SAC activity, Ca2+ responses to stretch and arterial contractions. Furthermore, by means of immunostainings and transmission electron microscopy, we observed that caveolae and caveolin-1 are expressed in PASMC from both Ctrl and CH rats and localize close to subplasmalemmal sarcoplasmic reticulum (ryanodine receptors) and mitochondria, thus facilitating Ca2+ exchanges, particularly in CH. In conclusion, caveolae are implicated in mechanotransduction in Ctrl PASMC by buffering mechanical forces. In PH-PASMC, caveolae form a distinct Ca2+ store facilitating Ca2+ coupling between SAC and sarcoplasmic reticulum.

Caveolae form Ï-structure plasma membrane invaginations of ~100 nm diameter, enriched in sphingolipids, cholesterol, and an oligomeric protein named caveolin. Caveolae are implicated in various cellular response mechanisms, like endocytosis, cholesterol homeostasis, and Ca2+ signaling. Owing to their composition, caveolae form stiff plasma membrane microdomains with emerging roles in mechanotransduction (25, 26). Mechanotransduction describes the ability of a cell to transduce a mechanical stimulus of stretch into a biological response, involving stretch-activated channels (SAC). A role in both mechanoprotection to membrane flattening (13, 23, 34) and mechanosensing to stretch has been ascribed to caveolae, depending on cell types (25). In fact, caveolae form dynamic plasma membrane microdomains importantly involved in the regulation of mechanical forces.

The caveolin protein (Cav) is essential for the formation of caveolae. Three isoforms of Cav have been identified and the one predominantly involved in the caveolae Ï-structure at the plasma membrane being Cav-1 (excepted in striated muscle where Cav-3 is predominantly responsible for the caveolae formation) (8). Deficient mice in Cav-1 lose caveolae at the plasma membrane and display some defects in lipid homeostasis in the adipose tissue, as well as some cardiovascular injury. Interestingly, these mice spontaneously develop pulmonary hypertension (PH) with right heart failure (38). PH is characterized by an increase of the mean pulmonary arterial pressure (PAPm) at rest over 25 mmHg (21). In human PH, PAPm at rest could be increased up to 90 mmHg in worst cases, whereas in normal subjects the value is around 14 mmHg (4, 21). In Cav-1–/– mice, the development of PH has been attributed to a defect in endothelium-dependent vasodilatation of intrapulmonary arteries (IPA) (39). Likewise in human, Austin et al. (4) recently showed that a mutation in the Cav-1 gene is correlated with some heritable forms of PH, which could be due to a defect in Cav-1 function in IPA endothelium. However, there is little information regarding the role of caveolae in the smooth muscle cell layer that constitutes the major part of the IPA wall. Pulmonary arterial smooth muscle cells (PASMC) are a main target of PH since they are involved in the remodeling and hyperreactivity of IPA, although the presence of caveolae and Cav-1 protein in PASMC is still a matter of debate (5, 10, 14, 16, 19, 20, 22, 27) and emerging hypotheses suggest that caveolae in PASMC may play a role during the establishment of PH (19) and in Ca2+ homeostasis in the disease (27).

During PH, mechanical forces exerted by the blood pressure on IPA wall are increased and in turn increase the vascular myogenic tone (11, 15), the main mechanism involved in mechanotransduction in vessels. In IPA, SAC allow a Ca2+ influx in PASMC and contribute to the myogenic tone (11, 15). We investigate here the involvement of caveolae in mechanotransduction in PASMC, under control and pathological conditions, i.e., PH induced in rats by a chronic hypoxia.

METHODS

Animal models. Wistar male rats were bred in agreement with the Local Animal Care Ethics Committee (Comité d’éthique de Bordeaux n° 50). The study was performed in accordance with the guidelines from Directive 2010/63/EU of the European Parliament on the protection of animals used for scientific purposes. Rats weighing 200–350 g were randomly assigned into two groups: control rats (Ctrl) were housed in ambient room air and chronically hypoxic rats (CH) rats were exposed 3 wk in a hypobaric chamber (50 kPa). For experiments, rats were euthanized with injection of pentobarbital sodium (190 mg/kg ip).
Immunostainings of IPA slides and in “en face.” Immediately after dissection, IPA from rats were fixed in 4% paraformaldehyde (PFA) for 15 min. After two washes in phosphate-buffered saline (PBS) solution, IPA were embedded in optimal cutting temperature compound and frozen sections of 10-μm thickness were obtained at −20°C by using a cryostat (Leica). After 45 min of incubating the slides in a blocking-permeabilized solution PTB 4% (4% BSA-0.1% Tween-20 in PBS solution, pH 7.4) at room temperature, slides were incubated overnight in PTB 1% with the Cav-1 [7C8] antibody (1:500; ab37141, abcam) at 4°C. Then, slides were washed three times in PBS solution and incubated during 2 h at room temperature with an Alexa-Fluor 568 goat anti-mouse antibody in PTB 1%. For controls, the primary antibody was omitted. After two washes in PBS solution, nuclei were stained in blue with DAPI for 10 min (1:500; Sigma-Aldrich). In sections, the internal elastic lamina has a green autofluorescence.

For “en face” stainings, IPA were immediately incubated overnight at 37°C in PTB 4% with the Cav-1 [7C8] antibody. The next day, IPA were washed four times in PBS solution and fixed during 15 min with PFA 4%. After two washes with PBS solution, IPA were incubated at 37°C during 3 h with the Alexa-Fluor 568 goat anti-mouse antibody in PTB 4%. For controls, the primary antibody was omitted. After two washes in PBS solution, nuclei were stained in blue with Hoechst 33342 for 10 min (1:500; Invitrogen). IPA were then longitudinally opened and observed under a Nikon D-Eclipse C1 confocal scanning microscope using a Nikon Apo Plan ×60/1.4 NA oil immersion objective. Fluorescent images were acquired and analyzed with the Nikon EZC1 software. In en face stainings, the internal elastic lamina (in green) allows to delimit smooth muscle cells from endothelial cells.

PASMC isolation, electrophysiological recordings, and microspectrofluorimetric measurements of [Ca2+]i. PASMCs were obtained by enzymatic dissociation of media, as previously described (15). Voltage-clamp recordings were made with a standard patch-clamp technique. Cells were bathed in a 310 mosM solution (in mM: 100 NaCl, 40 KCl, 2.2 CaCl2, 1.2 MgCl2, 14 glucose, 10 HEPES, pH = 7.4 adjusted with KOH). The recording pipette was filled with a 310 mosM solution (in mM: 137 NaCl, 5.4 KCl, 2.2 CaCl2, 1.2 MgCl2, 14
Caveolae and Pulmonary Hypertension

Fig. 2. Cav-1 expression in CH. **A**: Western blot for the Cav-1 of intrapulmonary arterial lysates from Ctrl and CH rats, normalized with the β-actin. A Mann-Whitney test was performed. *Significant difference for $P < 0.05$. There is a significantly decreased expression of Cav-1 in CH compared with Ctrl-IPA. **B**: nondenaturing Western blot showing multimers of Cav-1 at 250 and 465 kDa.

### RESULTS

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**Immunostainings in freshly isolated PASMC.** Five hours after their isolation, PASMC were fixed with PFA 4% for 15 min at 4°C. After two washes in PBS solution, cells were incubated 45 min with PTB 4% at room temperature. Cells were then incubated overnight in PTB 4% with the ryanodine receptors (RyR) 1 (1:200; ARR-001, Alomone Labs) or RyR3 antibodies (1:200; ARR-003, Alomone Labs), plus the Cav-1 [7C8] antibody (1:500; ab37141, abcam), at 4°C. The next day, cells were washed three times in PBS solution and incubated during 2 h at room temperature with an Alexa-Fluor 488 goat anti-mouse and Alexa-Fluor 568 goat anti-rabbit antibodies in PTB 4%. For controls, the primary antibody was omitted. After two washes in PBS, nuclei were stained in blue with DAPI for 5 min (1:500; Sigma-Aldrich).

*Western blot.* Western blots were performed as previously described (15). Briefly, 30 μg of homogenized IPA were loaded in a Tris-acetate 3–15% gel. After an overnight liquid transfer (4°C at 40 V) of the gel to a PVDF membrane, the membrane was saturated with 4% semiskimmed milk during 45 min. Then, membranes were incubated overnight at 4°C with the Cav-1 [7C8] antibody (1:1,000; ab37141, abcam) and washed three times with PBS-Tween 0.05% the next day. Membranes were subsequently incubated during 2 h at room temperature with a horseradish peroxidase-linked secondary antibody (1:10 000, Bio-Rad). After three washes in PBS-Tween 0.05%, the membrane was processed to a chemiluminescent detection (Substrat HR Immobilon Western, WBKLS0500; Millipore), according to the manufacturer’s instructions. Immunoblots were then revealed by enhanced chemiluminescence, acquired with a Bio-Rad camera.

### Statistical analysis

Results are expressed as means ± SE. Statistical analysis was performed on cells for patch-clamp experiments and Ca$^{2+}$ measurements (the number $n$ of tested responsive cells is indicated in each bar graph figure). For contraction experiments, the n indicated in each bar graph represent the number of arterial rings. For Western blots experiments, $N$ represent the number of rats. Each statistical test is indicated in figure legend. Statistical analysis was performed with the Prism software (GraphPad).

### Data and statistical analysis

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respectively (Fig. 1B). Because caveolae are membrane structures of less than 100 nm, transmission electron microscopy was also performed on freshly isolated PASMC to detect them. As shown in Fig. 1D, caveolae with a "U" shape are observed at the plasma membrane.

Cav-1 expression in pulmonary hypertensive IPA. Likewise, in CH rats, Cav-1 is expressed in SMC and EC of IPA in slide sections and in en face preparations (Fig. 6, A and B). However, Western blot experiments showed that expression of Cav-1 is decreased in IPA homogenates without endothelium from CH rats compared with Ctrl rats (Fig. 2, A and B). Besides, in a nondenaturing Western blot, multimers of Cav-1 are observed at 50 and 145 kDa (Fig. 2B), reflecting the formation of Cav-1 complexes, as observed in caveolae.

Caveolae participate to mechanosensing of PASMC. Next, we evaluate the involvement of caveolae in mechanosensing. In cell-attached configuration, SAC were activated by a negative pressure of −40 mmHg applied through the patch-clamp pipette on freshly isolated PASMC. Experiments were performed at −80 mV to measure inward currents, in the absence or in the presence of the caveolae disruptor: MβCD (5 mM). Whereas MβCD increased stretch-induced SAC activity in Ctrl-PASMC (Fig. 3, A and B), the opposite occurred in CH-PASMC, i.e., stretch-induced SAC activity was reduced by MβCD (Fig. 3, A and B). Besides, MβCD did not change the amplitude of the SAC unitary current (2.26 ± 0.07 pA vs. 2.28 ± 0.06 with MβCD).
Activation of SAC promotes an influx of Ca$^{2+}$ leading to an increase in the [Ca$^{2+}$], (15). Stretch-induced variation in [Ca$^{2+}$], was then measured in response to an osmotic shock of 225 mosM, another technique used to activate SAC (15), to freshly isolated PASMC, in the absence or presence of MβCD. We observed that MβCD did not modify the osmotic shock-induced Ca$^{2+}$ increase in Ctrl-PASMC whereas it significantly reduced that in CH-PASMC (Fig. 3, C–E). Besides, in Ctrl-PASMC (with or without MβCD) the osmotic shock-induced Ca$^{2+}$ increase was significantly inhibited by the SAC inhibitor GsMTx-4 and abolished in absence of extracellular Ca$^{2+}$ (Fig. 3F). Dantrolene, an inhibitor of RyR, also inhibited the response (Fig. 3F). On CH-PASMC, dantrolene reduced osmotic shock-induced Ca$^{2+}$ increase only in the absence of MβCD (Fig. 3G).

Caveolae participate in mechanodependent contraction of IPA. We further investigated the involvement of caveolae in the vascular tone by recording developed tension of IPA rings in response to an osmotic shock. Experiments were performed in the presence of L-NAME (100 μM) to block endothelium-induced vasodilatation. We observed in phenylephrine-precontracted IPA rings from Ctrl rats that an osmotic shock induces a contraction (Fig. 4A) and that this contraction was significantly greater in IPA rings from CH rats (Fig. 4, A and B). Furthermore, osmotic shock-induced contractions were increased by MβCD in Ctrl-IPA whereas they were reduced by MβCD in CH-IPA (Fig. 4B). These observations are in agreement with the increased SAC activity and osmotic shock-induced Ca$^{2+}$ increases observed in CH-PASMC compared with Ctrl-PASMC and the related reduced responses by MβCD observed in CH-PASMC (Fig. 3). Furthermore, osmotic shock-induced contractions were significantly inhibited by preincubation of IPA rings with different SAC inhibitors like GsMTx-4 or Gd$^{3+}$ (Fig. 4C).

We also performed osmotic shocks in the absence of extracellular Ca$^{2+}$. We observed that in the absence of extracellular Ca$^{2+}$ + 1 mM EGTA, an osmotic shock did not induce a contraction of Ctrl-IPA (Fig. 4, D and F). Conversely, in CH-IPA, a contraction is observed (Fig. 4, E and F). This is in agreement with previous results showing that osmotic shocks without extracellular Ca$^{2+}$ only provoke an increase of the [Ca$^{2+}$]i in CH-PASMC (15).

Caveolae act as a Ca$^{2+}$ store in PASMC. We previously shown that, in CH-PASMC, Ca$^{2+}$ responses without extracellular Ca$^{2+}$ involve caveolae since they are blocked by MβCD (15). To determine the origin of this Ca$^{2+}$ response, we simultaneously measured Ca$^{2+}$ by a confocal microscope in the sarcoplasmic reticulum (SR) and in mitochondria with the Ca$^{2+}$ probes fluo-5N and rhod-2, respectively. Figure 5A shows that, in the absence of extracellular Ca$^{2+} + 1$ mM...
EGTA, an osmotic shock decreases the [Ca\textsuperscript{2+}]\textsubscript{SR} and simultaneously increases the [Ca\textsuperscript{2+}]\textsubscript{m} in CH-PASMC. This finding suggests that at least a fraction of the cytoplasmic Ca\textsuperscript{2+} increase is due to a Ca\textsuperscript{2+} release from the SR. Moreover, in Ctrl and CH-PASMC, caveolae, subplasmalemmal SR, and mitochondria are closely located as shown by transmission electron microscopy, to facilitate Ca\textsuperscript{2+} exchanges (Fig. 5, B and C).

With respect to the Ca\textsuperscript{2+} channels of the SR implicated in osmotic shock-induced Ca\textsuperscript{2+} responses (15), immunostainings were performed between Cav-1 and RyRs in freshly isolated PASMC (Fig. 6C). We observed that Cav-1 is expressed at the plasma membrane and that RyR are located close to Cav-1, forming Ca\textsuperscript{2+} microdomains in cells (Fig. 6C). This is particularly obvious for RyR1, which is the main subtype expressed in the subplasmalemmal area in those cells (7, 15), RyR3 are expressed deeper in cells and distant to Cav-1 (Fig. 6C). Finally, we used the Duolink PLA technology that identifies nearby proteins and confirmed that Cav-1 is actually close to RyR1 in the subplasmalemmal of PASMC from Ctrl and CH rats (Fig. 6D). By quantifying fluorescence spots, we observed that there is significantly more Cav-1 close to RyR1 in CH-PASMC rather in Ctrl (Fig. 6D). To confirm that SAC could be located in the caveolar microenvironment, we performed a Duolink experiment between the putative SAC TRPV4 and Cav-1. TRPV4 is a SAC channel and we previously shown that this channel participates to the Ca\textsuperscript{2+} increase induced by an osmotic shock in PASMC (24). As observed in Fig. 6E, TRPV4 are actually close to Cav-1, in both Ctrl and CH-PASMC.

This way, even if the distance between caveolae and subplasmalemmal SR is not different between Ctrl and CH-PASMC (Fig. 5C), Fig. 6D shows that RyR1 are closer to caveolin-1. To address whether the coupling between caveolae and the SR could be functionally higher in CH-PASMC, we measured osmotic shock-induced Ca\textsuperscript{2+} increases in the presence of M\textbeta\textsuperscript{CD} and a RyR1/3 inhibitor, dantrolene. In Ctrl-PASMC, in both conditions (without and with M\textbeta\textsuperscript{CD}), Ca\textsuperscript{2+} responses to an osmotic shock are significantly decreased in the presence of dantrolene (10 \textmu M) (Fig. 3F). Conversely, in CH-PASMC, dantrolene inhibited the osmotic shock-induced Ca\textsuperscript{2+} increases without M\textbeta\textsuperscript{CD} but not with M\textbeta\textsuperscript{CD} (Fig. 3G).

**DISCUSSION**

The present study has examined the role of caveolae in mechanotransduction in the pathophysiology of pulmonary arteries. Our major finding is that, in PH-PASMC, caveolae may form a distinct Ca\textsuperscript{2+} store facilitating Ca\textsuperscript{2+} coupling between SAC and SR, implicating RyR1 (Fig. 7).

Using transmission electron microscopy, we have shown that caveolae are present in native smooth muscle cells, freshly isolated from rat IPA. These results are in agreement with previous studies revealing the presence of caveolae in cultured PASMC (27). Nevertheless, in the literature, there remain some discrepancies regarding the presence of the caveolae-1 protein in PASMC, due to immunostaining acquisitions with a poor resolution (5, 10, 19). Here, using confocal microscopy imaging with an adequate resolution, we showed that the Cav-1 protein is expressed in both EC and SMC of rat IPA, in slides, in en face preparations, and also on freshly isolated PASMC (Fig. 1). Furthermore, Western blot data obtained in IPA without endothelium support this finding (Fig. 1B).

Caveolae have been implicated in shear stress-induced vasodilation in EC (30–32, 35) acting on both plasma membrane fluidity within caveolae and caveolae-associated proteins (TRP channels, eNOS) (3). In the arterial wall, SMC are not exposed to shear stress because of their location under the internal elastic lamina. Nevertheless, they are subjected to transmural wall pressure variations exerted by the blood pressure. We thus studied the role of caveolae in the response to membrane stretch (applied by a negative pressure or by an...
osmotic shock) in PASMC from Ctrl rats. At level of caveolae, the plasma membrane is stiffer and less elastic (12). If SAC are localized into these lipid rafts, the pressure will have a modest impact on their activity (17). In the presence of M/H9252CD, the stiffness of the plasma membrane will decrease, changing the properties of the plasma membrane microenvironment surrounding SAC. Thus, as the plasma membrane is smoother with M/H9252CD, in response to an identical stretch the activity of SAC will increase, as observed (Fig. 3B). As a consequence, Ca²⁺ influx through SAC and hence, the resulting contraction response will increase too (Figs. 3B and 4B). Thereby, under physiological conditions, caveolae play a protective role in IPA that slows down mechanotransduction. Some hypotheses in the literature suggest that caveolae may form a quickly available reserve of plasma membrane that could participate to cell membrane plasticity in response to a mechanical stress (23, 25, 34). Collectively, caveolae may act as plasma membrane buffers to support mechanical stress while controlling the membrane plasticity, SAC activity and allowing IPA to finely adapt its tone to the pressure.

During PH, the pressure increase in IPA (28) and thus stretch forces exerted on PASMC also increase. As previously shown, stretch-induced SAC activity and the resulting Ca²⁺ increase in response to an osmotic shock are increased in PASMC from CH rats (11, 15). In the present study, we further demonstrate that an osmotic shock is able to induce a contrac-
tion of the IPA. These osmotic shock-induced contractions are enhanced in CH-IPA, supporting the hyperreactivity of IPA to stretch, as previously observed for the myogenic tone, which is increased in CH-IPA (11). However, in contrast to what occurs in Ctrl rats, disrupting caveolae with MβCD reduces SAC activity, Ca^{2+}, and contraction responses to stretch in IPA from CH rats. These findings suggest that caveolae play an important role in mechanosensing in PASMC and, particularly, in pulmonary hypertensive rats. In the systemic circulation, previous studies have shown that caveolae are involved in the myogenic tone (1, 2, 29). Yet we describe here a distinct role of caveolae under a pathological condition, in a vascular system highly sensitive to an increased pressure: the pulmonary hypertensive IPA.

Three hypotheses may be put forward to account for such distinct role of caveolae in pulmonary hypertensive IPA. Firstly, we observed a decreased expression of Cav-1 (both monomers and multimers) in CH-IPA (Figs. 2B and 6, A and B). If there are fewer caveolae, the plasma membrane will be less stiff and thus more sensitive to stretch forces, explaining at least in part why SAC activity, Ca^{2+}, and contraction responses to stretch are increased in CH compared with Ctrl rats. Previous studies (18, 19, 27, 36) have reported conflicting results about the variation of expression of Cav-1 during pulmonary hypertension. These differences could be due to (1) the type of pulmonary hypertension considered (animal model, human model), (2) the type of tissues used in the experimental procedures (whole lung, isolated pulmonary artery with or without endothelium), or (3) the type of pulmonary hypertension. Secondly, in PASMC from PH rats, caveolae seem to be involved in strengthening the coupling between SAC and subplasmalemmal RyR1, thus enhancing the Ca^{2+}-induced Ca^{2+}-release (CICR) mechanism that we previously described in PASMC (15). To support this hypothesis, using both immunostainings and Duolink PLA technology, we evidenced that Cav-1 are close to RyR1 and this coupling is reinforced in CH-PASMC (Fig. 6D). This organization between plasma membrane caveolae and subplasmalemmal Ca^{2+} stores may contribute to form specialized Ca^{2+} microdomains in PASMC, also in connection with mitochondria. Besides, in Fig. 3F, we shown that, in Ctrl-PASMC, MβCD has no effect on the inhibition % of the Ca^{2+} response in dantrolene. This suggests that, in Ctrl-PASMC, caveolae are not implicated in the CICR between SAC and RyR1. Conversely, in CH-PASMC, dantrolene inhibits the osmotic shock-induced Ca^{2+} increases without MβCD but not with MβCD (Fig. 3G). Altogether, this suggests that, in Ctrl-PASMC, caveolae are not involved in the CICR mechanism but rather act on the SAC activity (Fig. 3B). In CH-PASMC, caveolae are important for the CICR mechanism, especially thanks to a stronger coupling between RyR1 and Cav-1 (Fig. 6D).

Thirdly, in a former study, Haack et al. (16) used pyroantimicrobial granules associated to transmission electron microscopy to visualize the localization of Ca^{2+} ions in cell microdomains on PASMC from both normal and CH rats. Interestingly, they observed that in Ctrl rats caveolae are present in PASMC but do not contain Ca^{2+}, whereas on PASMC from CH rats caveolae accumulate Ca^{2+} ions (16). Later, in 2006, Daniel et al. (9) formulated the hypothesis that caveolae in smooth muscle cells could form a separated Ca^{2+} store where Ca^{2+} is concentrated and does not equilibrate with the extracellular medium. According to our present results, this hypothesis could apply to CH rats exclusively, i.e., caveolae accumulating Ca^{2+} to act as a specific Ca^{2+} store. To support this hypothesis, the plasma membrane calcium ATPase pumps (PMCA) or the Na^{+}/Ca^{2+} exchanger whose expression is increased during PH (37) could be located within caveolae in rats, as previously shown in bovine PASMC (14, 33), and contribute to refill caveolae in Ca^{2+}. Under such conditions, in the absence of Ca^{2+} in the bathing solution, Ca^{2+} ions may still be present into caveolae and participate to the Ca^{2+} increase and the contraction response induced by an osmotic shock, as observed.
To conclude, whereas caveolae in endothelial cells were previously shown to be involved in PH, we showed here that they may also play an important role in SMC. In this connection, even if Cav-1 expression in endothelium is reduced in IPA from PH patients such is not the case in SMCs (4), suggesting that caveolae in the SMC layer of IPA could play an important role in this disease, separately from their role in the endothelium. Finally, our study highlights a role of caveolae in mechanotransduction in the pathophysiology of pulmonary arteries. In control IPA, caveolae have a mechanoprotective role and confer to the plasma membrane a plasticity to restrain mechanotransduction. However, under conditions in which mechanical forces are increased such as PH, caveolae are implicated in a modification of the coupling between the stretch stimulus and intracellular Ca2+ signaling pathways.

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DISCLOSURES

No conflicts of interest, financial or otherwise, are declared by the author(s).

AUTHOR CONTRIBUTIONS


REFERENCES


