Activation of ryanodine receptor/Ca^{2+} release channels downregulates CD38 in the Namalwa B lymphoma

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Abstract CD38 is a multifunctional ectoenzyme that catalyses formation of cyclic ADP ribose (cADPr), a second messenger that opens ryanodine receptor (RyR) Ca^{2+} channels. Despite its importance in signal transduction processes, little is known about the mechanisms regulating CD38 expression levels. In the current study, ryanodine stimulation of Ca^{2+} release in Namalwa cells decreased both CD38 protein abundance and cyclase activity. Reductions in cyclase activity were prevented by RyR antagonists, by lysosomal blockers, though not by calpain or proteasomal inhibitors. These findings indicate a novel negative feedback mechanism between RyR channel activity and CD38 abundance acts in cADPr signal transduction.

Key words: CD38; Lysosome; Ca^{2+}; Cyclic ADP ribose; Ryanodine receptor

1. Introduction

CD38 is a multifunctional ectoenzyme expressed in a wide range of cell types, including lymphocytes, pancreatic β cells, neurones and smooth muscle cells [1]. This enzyme is a type II plasma membrane protein that catalyses formation of cyclic ADP ribose (cADPr) from extracellular nicotinamide adenine dinucleotide (NAD). Dimerisation of CD38 [2] and/or internalisation of membrane vesicles bearing this enzyme [3,4] facilitates entry of cADPr into the cytoplasm, where this cyclic nucleotide acts as a second messenger, opening ryanodine receptor (RyR)/Ca^{2+} release channels in the endoplasmic reticulum, thereby increasing cytosolic free Ca^{2+} $[Ca^{2+}]_c$. Elevations in $[Ca^{2+}]_c$ subsequently regulate a plethora of cellular processes, including proliferation, gene expression, secretion, motility, necrosis and apoptosis [5]. Despite its significance in signal transduction processes, little is known about mechanisms regulating CD38 protein abundance. Interleukin-4 downregulates CD38 in a human B cell line, via a serine/threonine protein kinase-dependent pathway [6]. In renal mesangial cells, CD38 is upregulated by tumour necrosis factor-α, interleukin-1β and all-*trans*-retinoic acid [7]. In a variety of cell types, activation of CD38 by NAD, or by antibody-mediated cross-linking, decreases surface expression of the cyclase by a mechanism dependent on membrane internalisation [3,4,8,20].

In mammals, the RyR family consists of three isoforms of cation channel that release Ca^{2+} from intracellular stores, such as the endoplasmic and sarcoplasmic reticulum. Although these channel proteins were originally characterised in tissues where they are present at high levels, namely skeletal muscle (RyR1) and heart (RyR2), they are also expressed in a broad range of non-muscle cells [9–12,28]. Despite being pivotal in striated muscle excitation–contraction coupling, the roles of RyRs in other cell types have not been extensively defined. B lymphocyte cell lines and CD19+ B cells isolated from blood express functional RyR1 channels that are activated in response to B cell receptor cross-linking [12]. Pharmacological activation of B cell RyRs using the agonists 4-chloro-*meta*-cresol (CmC) or caffeine increases interleukin-1β secretion [13], indicating that Ca^{2+} signalling via these channels plays a role in regulation of immune response.

Namalwa Burkitt's lymphoma, a CD19+ model of the mature human B cell, expresses CD38 and has been used extensively in the investigation of the catalytic properties of this cyclase [3]. In spite of this, little is known about the relationships between CD38 and RyR Ca^{2+} signalling in this B lymphocyte cell line. In the current study, the presence of functional RyRs in Namalwa cells and the biological consequences of their prolonged activation were analysed [14]. In particular, the relationships between CD38 activity and RyR-mediated Ca^{2+} release were investigated. For the first time, it was demonstrated that: (1) Namalwa B cells express the RyR1 isoform, which mobilises Ca^{2+} in response to millimolar CmC or nanomolar ryanodine; (2) prolonged (24 h) stimulation of Namalwa cells with ryanodine results in a dramatic reduction in CD38 protein level, GDP ribosyl cyclase activity and Ca^{2+} mobilisation elicited by extracellular NAD; (3) these effects are specific, since levels of surface IgM μ chain are not influenced by ryanodine treatment; (4) CD38 downregulation is abrogated by procaine or dantrolene, RyR antagonists; and (5) RyR-mediated CD38 downregulation is dependent on the lysosomal pathway of proteolysis, since this effect is blocked by pretreatment with ammonium chloride or chloroquine, though not by calpain and proteasome inhibitors. These observations indicate a novel negative feedback mechanism operates in B lymphocyte CD38 Ca^{2+} signalling: prolonged ac-

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Abbreviations: ALLM, *N*-acetyl-Leu-Leu-Met-al; cADPr, cyclic ADP ribose; cGDPr, cyclic GDP ribose; CmC, 4-chloro-*meta*-cresol; Fluo-3AM, fluo-3-acetoxymethyl ester; KHB, Krebs–Henseleit buffer; MG132, *Z*-Leu-Leu-Leucinal; NAD, nicotinamide adenine dinucleotide; NGD, nicotinamide guanine dinucleotide; RT-PCR, reverse transcription polymerase chain reaction; RyR, ryanodine receptor

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tivation of RyRs resulting in lysosomal downregulation of the cADPr producing ectoenzyme.

2. Materials and methods

2.1. Materials

A549 lung epithelial carcinoma, Jurkat T lymphoma, Namalwa Burkitt's lymphoma and SHSY5Y neuroblastoma cell lines were cultured according to the protocols of the European Collection of Animal Cell Cultures. Ryanodine, CmC, human IgM, anti-human IgM μ chain peroxidase conjugate and affinity-purified goat anti-human IgM μ chain were from Merck. Mouse anti-CD38 monoclonal antibody AT1 was purchased from Santa Cruz Biotech. Fluo-3-acetoxymethyl ester (Fluo-3AM) was from Molecular Probes. *Aplysia californica* ribosyl cyclase, NAD, nicotinamide guanine dinucleotide (NGD) and protease inhibitors were from Sigma-Aldrich. SuperSignal Enhanced Chemiluminescence reagents were from Pierce. All other reagents were of analytical grade or better.

2.2. Measurement of $[Ca^{2+}]_c$ using Fluo-3

Namalwa B cells were harvested by centrifugation at $1000\times g$ for 5 min, washed once with Krebs–Henseleit buffer (KHB: 120 mM NaCl, 4.8 mM KCl, 1.2 mM $MgSO_4$, 1.2 mM KH_2PO_4, 25 mM $NaHCO_3$, 10 mM glucose, 1 mM probenecid, 5 mM HEPES pH 7.2) containing 2 mM $CaCl_2$, then they were resuspended at a density of 6×10^6 cells/ml in KHB containing 2 mM $CaCl_2$ and 5 μM of Fluo-3AM. Following incubation for 1 h at room temperature in the dark, Namalwa cells were washed twice with KHB, then they were resuspended in 3 ml of KHB (2×10^6 cells/ml) containing 2 mM $CaCl_2$, or in some cases 0.5 mM EGTA, in a stirred fluorimetry cuvette at 37°C. Fluo-3 fluorescence (490 nm excitation/530 nm emission) was monitored using either a Perkin-Elmer LS50B or a Wallac Varian Eclipse luminescence spectrometer. At the end of all experiments, fluorescence measurements were calibrated to $[Ca^{2+}]_c$ levels by addition of 1% Triton X-100, then 4 mM EGTA, according to the protocol of Grynkiewicz et al. [15].

2.3. Miscellaneous

Reverse transcription polymerase chain reaction (RT-PCR) was employed to determine the RyR isoform expression profile of Namalwa cells, using the protocol of Sei et al. [12], as modified by Hosoi et al. [16]. The NGD cyclase activity of Namalwa cells was assayed using the protocol of Graeff et al. [17]. In brief, cells (0.5×10^6) were lysed in 500 μl of 20 mM Tris–HCl pH 7.4 by five passes through a 19 gauge needle, then were incubated with 100 μM of NGD for 1 h at 37°C. Reactions were stopped by addition of 10 μl of 50% trichloroacetic acid and the fluorescent product (cyclic GDP ribose, cGDPr) measured at excitation/emission wavelengths of 300 nm/410 nm. Concentrations of cGDPr were determined from fluorescence values using a standard curve generated using known amounts of cGDPr, produced using *A. californica* ADP ribosyl cyclase. For protease inhibition experiments, cells were pretreated with *N*-acetyl-Leu-Leu-Met-al (ALLM), calpain inhibitor II, chloroquine or *Z*-Leu-Leu-Leucinal (MG132) for 30 min, then were pelleted and resuspended in fresh culture medium without inhibitor prior to 24 h incubation with ryanodine. This was necessary because prolonged treatment of Namalwa lymphocytes with these inhibitors significantly increased cell death. Other inhibitors were added at the same time as ryanodine and co-incubated for 24 h. Protein concentrations were determined using the method of Bradford [19], using bovine serum albumin as a standard. Numerical data are expressed as mean values ± S.E.M. and n was the number of experiments performed. Data were fitted to the Hill equation using GraphPad Prism software. Differences between samples were analysed using a paired Student's t-test using Microsoft Excel.

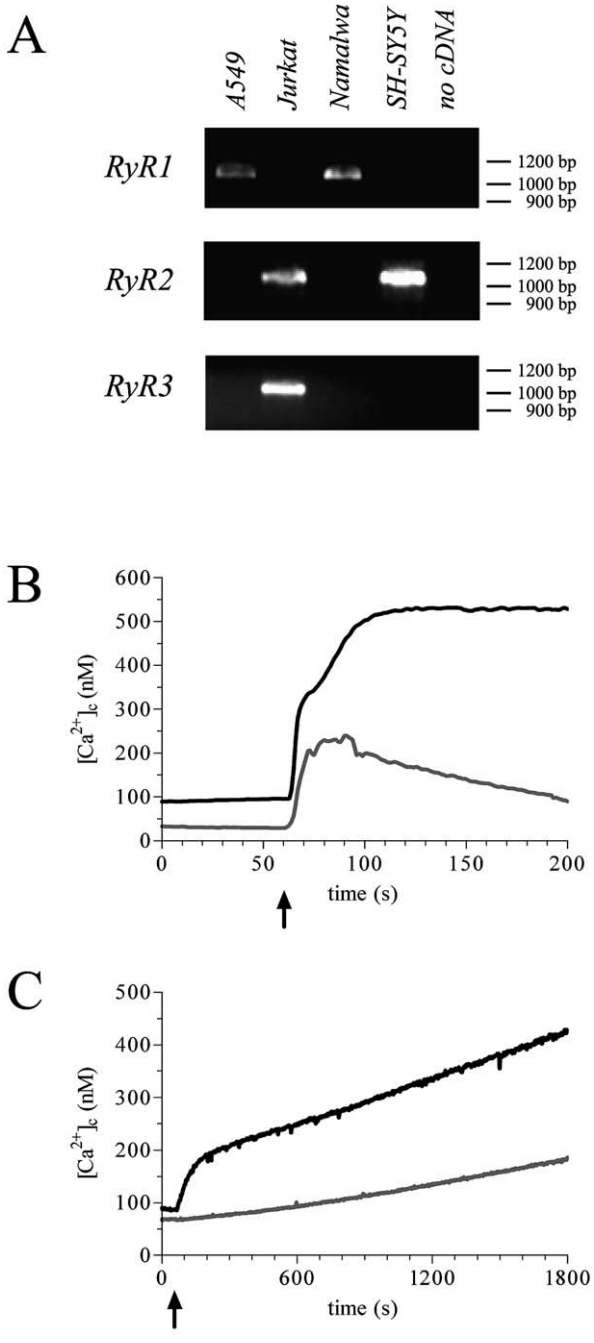

Fig. 1. Namalwa B lymphoma cells express functional RyR1 Ca^{2+} channels. A: Namalwa cells express RyR1 mRNA. Shown are agarose electrophoresis gels of PCR products amplified from cDNA from various cell lines using RyR isoform-specific primer sets. PCR products formed are 1112 bp for RyR1, 1083 bp for RyR2 and 1015 bp for RyR3. B: The RyR agonist CmC increases $[Ca^{2+}]_c$ in Namalwa cells by releasing this ion from intracellular stores. In the presence of 2 mM extracellular Ca^{2+} (black trace), addition of 5 mM CmC (arrow) increased $[Ca^{2+}]_c$ from a resting level of 83 ± 5 nM (n = 25) to a peak of 535 ± 56 nM (n = 5). In nominally Ca^{2+}-free extracellular medium (0.5 mM extracellular EGTA, grey line), 5 mM CmC elevated $[Ca^{2+}]_c$ from 39 ± 4 nM (n = 5) to a maximum of 207 ± 34 nM (n = 5). C: Ryanodine causes a sustained increase in $[Ca^{2+}]_c$ in Namalwa B cells that can be blocked with the RyR antagonist procaine. Addition of 100 nM ryanodine (arrow) to cells (2 mM $CaCl_2$ in medium) increased $[Ca^{2+}]_c$ from 91 ± 8 nM (n = 3) at rest to a peak of 217 ± 14 nM (n = 3) after 5 min of ryanodine addition. Preincubation of the cells with 10 mM procaine, a RyR antagonist, significantly ($P < 0.005$) reduced this maximum elevation in $[Ca^{2+}]_c$ to 115 ± 9 nM (n = 3) after 5 min of ryanodine addition, from a basal level of 71 ± 11 nM (n = 3).

3. Results and discussion

Namalwa Burkitt's lymphoma, a model of the mature human B lymphocyte, expresses RyR/Ca^{2+} release channels. Efforts to identify the subtype(s) of RyR expressed in Namalwa cells by probing Western blots of cell lysates with a range of isoform-selective antisera [9,10] were unsuccessful, possibly as a result of the very low relative abundance of these proteins in B lymphocytes [12]. Consequently, RyR isoform-specific RT-PCR [19] was employed to identify the subtype(s) of RyR expressed in Namalwa cells. Using this technique it was found that like A549 epithelial cells [28], Namalwa lymphocytes express detectable levels of RyR1 mRNA only (Fig. 1A), in contrast to Jurkat T cells that possess RyR2 and RyR3 [12], and SHSY5Y neuroblastoma cells that have RyR2 [9]. This observation is discrepant from data presented by Hosoi et al. [16], who report that Namalwa cells do not express any RyR mRNA. The reasons for this difference are unclear, although optimal $MgCl_2$ concentrations for amplification of RyR cDNA in our laboratory were distinct from those employed by Sei et al. [12]. Type 1 RyR expressed in Namalwa B lymphocytes forms functional Ca^{2+} release channels. Stimulation of these cells with a maximal concentration (5 mM) of the RyR agonist CmC leads to an increase in $[Ca^{2+}]_c$ that persists in EGTA-buffered extracellular medium (Fig. 1B), indicating that this ion is released from intracellular stores. Namalwa cell $[Ca^{2+}]_c$ responses to CmC are dose-dependent, displaying a half-maximal effective concentration of 0.7 ± 0.1 mM ($n = 3$)

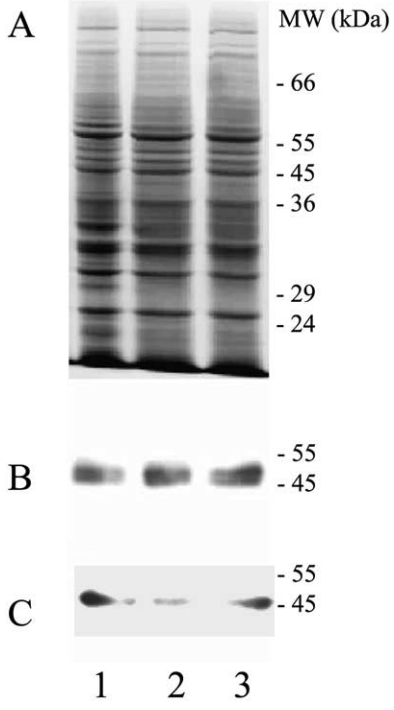

Fig. 2. Chronic stimulation of Namalwa B cells with ryanodine leads to downregulation of the cADPr-generating enzyme CD38. Namalwa cells were treated for 24 h with 1% DMSO (lane 1), 100 nM ryanodine (lane 2), or 100 nM ryanodine plus 1 mM procaine (lane 3). Cell lysates (50 μg protein/lane) were then resolved on 10% reducing sodium dodecyl sulfate–polyacrylamide gel electrophoresis, then either (A) stained with Coomassie R250 or transferred onto nitrocellulose and immunostained with (B) 1:1000 anti-human IgM μ chain or (C) 5 μg/ml anti-CD38 monoclonal antibody AT1. Data shown are representative of three independent experiments.

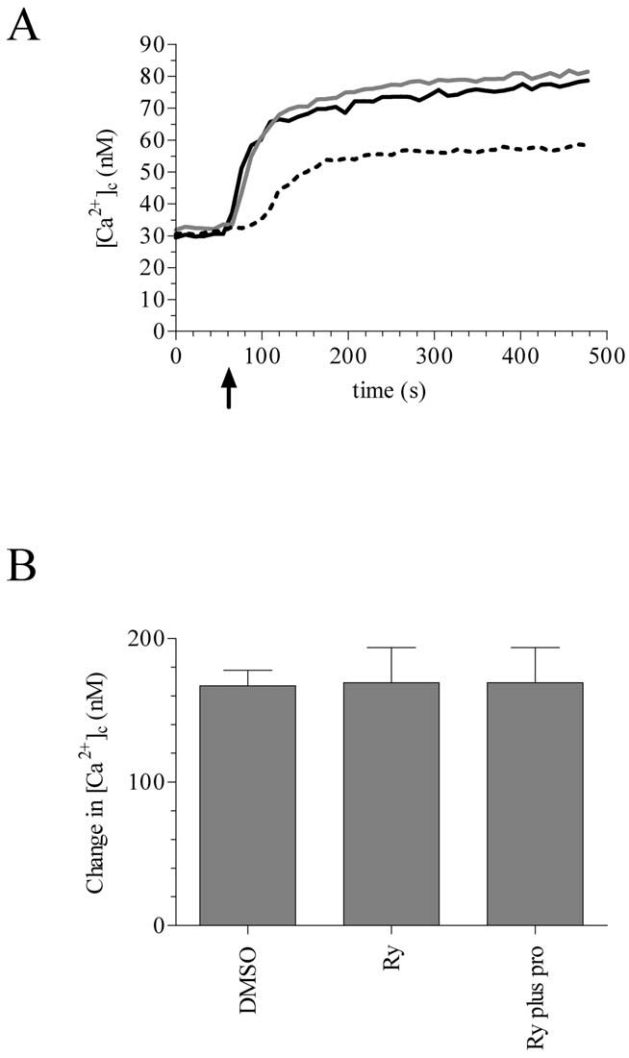

Fig. 3. Chronic ryanodine stimulation of Namalwa cells results in a decrease in $[Ca^{2+}]_c$ rises triggered by extracellular NAD. A: Following incubation with either 1% DMSO (solid black trace), 100 nM ryanodine (broken black line) or 100 nM ryanodine plus 1 mM procaine (solid grey line), Namalwa cell $[Ca^{2+}]_c$ responses to extracellular NAD (5 mM, arrow) were monitored under nominally Ca^{2+}-free conditions. Chronic ryanodine stimulation significantly ($P < 0.01$) reduced $[Ca^{2+}]_c$ responses to extracellular NAD, from a peak change in $[Ca^{2+}]_c$ of 56 ± 5 nM in DMSO-pretreated cells to 29.1 ± 0.5 nM in ryanodine-pretreated lymphocytes. Procaine prevented this decrease, with NAD causing a change of $[Ca^{2+}]_c$ of 51 ± 4 nM in ryanodine-plus procaine-pretreated cells ($n = 3$ in all cases). B: Long-term (24 h) treatment of Namalwa cells with 1% DMSO ('DMSO'), 100 nM ryanodine ('Ry'), or 100 nM ryanodine plus 1 mM procaine ('Ry plus pro') did not alter their responsiveness to CmC. Shown are the peak $[Ca^{2+}]_c$ rises in these three groups following stimulation with 1 mM CmC ($n = 4$ in all cases).

and a Hill slope of 2.7 ± 0.2 ($n = 3$). These parameters are similar to those reported for CmC-induced Ca^{2+} responses in normal Epstein–Barr virus-immortalised human B cells [13]. Prolonged exposure (> 60 min) of Namalwa cells to CmC concentrations greater than 2 mM resulted in a significant increase in the number of trypan blue-permeable cells that was independent of Ca^{2+} mobilisation, since it could not be blocked by the RyR antagonists dantrolene or procaine (data not shown). Consequently, in order to examine the consequences of long-term RyR activation in Namalwa

B lymphocytes, cells were stimulated with 100 nM ryanodine, which had no significant effect on viability over a 24 h period. Ryanodine (100 nM) triggered a sustained increase in $[Ca^{2+}]_c$ that could be blocked by preincubation with procaine (Fig. 1C).

Prolonged (24 h) incubation of Namalwa B lymphocytes with 100 nM ryanodine had no significant effect on cell viability, proliferation, amount of total cellular protein, or quantity of secreted protein (data not shown). However, 24 h ryanodine stimulation drastically reduced CD38 abundance in Namalwa cells (Fig. 2C), despite having no detectable effect on surface IgM μ chain levels (Fig. 2B). Downregulation of CD38 could be abrogated using the RyR antagonist procaine (Fig. 2C). Since CD38 is capable of synthesising the Ca^{2+}-mobilising second messenger cADPr from NAD, it was anticipated that downregulation of this enzyme by chronic RyR activation would inhibit $[Ca^{2+}]_c$ rises stimulated by extracellular NAD. As expected, 24 h ryanodine treatment significantly decreased Namalwa B cell responses to NAD, this effect being prevented by coincubation with the RyR antagonist procaine (Fig. 3A). These findings are not a consequence of ryanodine or procaine remaining within cells following 24 h exposure and washing steps, or of other alterations in RyR function, since there were no significant differences between dimethyl sulphoxide (DMSO)-treated, ryanodine-treated, or ryanodine- plus procaine-treated Namalwa cells in the magnitude of $[Ca^{2+}]_c$ rises triggered by 1 mM CmC (Fig. 3B). This indicates that CD38 downregulation by chronic ryanodine treatment has direct negative impact on Ca^{2+} signalling in Namalwa cells, rather than being a consequence of modified RyR function.

To quantify CD38 activity following prolonged RyR stimulation, formation of fluorescent cGDPr from non-fluorescent NGD was monitored. Using this assay, it was found that ryanodine treatment had a half-maximal effect on NGD ribosyl cyclase activity in Namalwa cells after 3.9 ± 0.8 h ($n = 3$) and was complete ($40 \pm 5\%$ of original activity, $n = 3$) after $\sim$18 h. Consequently, cyclase assays were performed 24 h after ryanodine treatment. The GDP ribosyl cyclase assay was used to investigate the mechanisms responsible for CD38 downregulation in ryanodine-treated Namalwa cells. Procaine and the structurally unrelated RyR antagonist dantrolene blocked ryanodine-induced loss of cyclase activity (Table 1). Inhibitors of the calpain family of Ca^{2+}-dependent cysteine proteases, ALLM and calpain inhibitor II, could not prevent reduction in Namalwa NGD cyclase activity caused by ryanodine. Analysis of the primary structure of human CD38 using the PESTfind algorithm (http://www.at.embnet.org/embnet/tools/bio/PESTfind/) supports this observation, since no strong candidate calpain cleavage sites [18] were detected in this protein. Pretreatment of Namalwa cells with the proteasome inhibitor MG132 had no discernible effect on loss of CD38 activity. However, lysosomotropic agents blocked ryanodine-stimulated loss of CD38 in Namalwa lymphocytes, since there was no significant difference in cyclase activity between cells treated with ammonium chloride or chloroquine and cells treated with these compounds plus ryanodine (Table 1).

The data reported here indicate that prolonged incubation of Namalwa cells with ryanodine, at concentrations that cause sustained Ca^{2+} release via RyRs, downregulates CD38 protein via the lysosomal degradative pathway. Since CD38 is the key enzyme in mammalian cells responsible for the synthesis of cADPr [1], an RyR-activating second messenger, its downregulation represents a potential negative feedback mechanism in Ca^{2+} signal transduction processes. In this report, it is demonstrated for the first time that RyR activity can drive lysosomal degradation of CD38, since ryanodine-stimulated increases in $[Ca^{2+}]_c$ precede any reduction in the activity or abundance of this enzyme. Furthermore, Namalwa cell $[Ca^{2+}]_c$ elevations in response to extracellular NAD are diminished following chronic ryanodine stimulation, without any detectable decrease in RyR function. Proteolytic steps in negative feedback mechanisms that regulate Ca^{2+} signal transduction processes are not unprecedented. Chronic stimulation of a variety of cell types with hormones that cause sustained elevations in cytosolic inositol 1,4,5-trisphosphate levels triggers degradation of inositol 1,4,5-trisphosphate receptor/Ca^{2+} release channel proteins by the ubiquitin/proteasomal pathway [21,22]. RyRs are potentially regulated by this degradative system, since they are associated with a proteasomal recognition subunit S5a [23].

Table 1
Effect of protease inhibitors or RyR antagonists on Namalwa NGD ribosyl cyclase activity following chronic ryanodine stimulation

Treatment	Cyclase activity (nmol cGDPr formed/mg protein/min ± S.E.M.)	*P* versus no ryanodine ('−' vs. '+')	*n*
DMSO −	1.62 (±0.19)		4
DMSO +	0.64 (±0.07)	0.003	4
50 μM dantrolene −	1.65 (±0.11)		3
50 μM dantrolene +	1.56 (±0.18)	NS	3
1 mM procaine −	1.32 (±0.09)		3
1 mM procaine +	1.38 (±0.04)	NS	3
100 μM chloroquine −	1.46 (±0.17)		3
100 μM chloroquine +	1.60 (±0.21)	NS	3
10 mM NH_4Cl −	1.21 (±0.10)		4
10 mM NH_4Cl +	1.47 (±0.08)	NS	4
50 μM ALLM −	1.39 (±0.18)		3
50 μM ALLM +	0.67 (±0.13)	0.031	3
50 μM calpain inhibitor II −	1.33 (±0.10)		3
50 μM calpain inhibitor II +	0.48 (±0.05)	0.001	3
50 μM MG132 −	1.97 (±0.19)		3
50 μM MG132 +	1.01 (±0.08)	0.010	3

Namalwa cells were treated for 24 h in the presence ('+') or absence ('−') of 100 nM ryanodine plus protease inhibitors or RyR antagonists. Cellular NGD ribosyl cyclase activity was then measured as described in Section 2. Differences between ryanodine-treated and untreated activities were compared using a paired Student's *t*-test. NS, not significantly different, $P > 0.05$.

Several candidate mechanisms might underlie enhancement of lysosomal degradation of CD38 by RyR-mediated increases in $[Ca^{2+}]_c$. Endosome fusion is a Ca^{2+}-dependent event, regulated by the Ca^{2+} binding protein calmodulin [24]. Membrane fusion events are stimulated by inositol 1,4,5-trisphosphate receptor-mediated Ca^{2+} release from intracellular stores [25]. By analogy, RyR channel opening might promote endosome/lysosome fusion, thereby increasing proteolysis of proteins degraded via the lysosomal pathway. Activation of RyRs also stimulates translocation of membrane vesicles, as observed in the transport of aquaporin 2-bearing membranes triggered by vasopressin in inner medulla collecting duct cells [26]. Finally, 17β-estradiol causes an increase in $[Ca^{2+}]_c$ in mollusc blood cells. This $[Ca^{2+}]_c$ elevation promotes translocation of Ca^{2+}-dependent phospholipase A2 from the cytosol to membranes, which leads to an increase in lysosomal volume and proteolytic activity [27]. Whatever the basis of enhanced lysosomal turnover of CD38 by RyR activation, this process potentially represents a key negative feedback pathway in cADPr/Ca^{2+} signal transduction.

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References

[1] Lee, H.C. (1997) Physiol. Rev. 77, 1133–1164.
[2] Franco, L., Guida, L., Bruzzone, S., Zocchi, E., Usai, C. and De Flora, A. (1998) FASEB J. 12, 1507–1520.
[3] Zocchi, E., Franco, L., Guida, L., Piccini, D., Tacchetti, C. and De Flora, A. (1996) FEBS Lett. 396, 327–332.
[4] Zocchi, E., Usai, C., Guida, L., Franco, L., Bruzzone, S., Passalacqua, M. and De Flora, A. (1999) FASEB J. 13, 273–283.
[5] Berridge, M.J., Lipp, P. and Bootman, M.D. (2000) Nat. Rev. Mol. Cell Biol. 1, 11–21.
[6] Shubinsky, G. and Schlesinger, M. (1996) Cell. Immunol. 173, 87–95.
[7] Yusufi, A.N., Cheng, J., Thompson, M.A., Dousa, T.P., Warner, G.M., Walker, H.J. and Grande, J.P. (2001) Am. J. Physiol. Renal Physiol. 281, F91–F102.
[8] Pfister, M., Ogilvie, A., da Silva, C.P., Grahnert, A., Guse, A.H. and Hauschildt, S. (2001) Eur. J. Biochem. 268, 5601–5608.
[9] Mackrill, J.J., Challiss, R.A., O'Connell, D.A., Lai, F.A. and Nahorski, S.R. (1997) Biochem. J. 327, 251–258.
[10] Giannini, G., Conti, A., Mammarella, S., Scrobogna, M. and Sorrentino, V. (1995) J. Cell Biol. 128, 893–904.
[11] Bennett, D.L., Cheek, T.R., Berridge, M.J., De Smedt, H., Parys, J.B., Missiaen, L. and Bootman, M.D. (1996) J. Biol. Chem. 271, 6356–6362.
[12] Sei, Y., Gallagher, K.L. and Basile, A.S. (1999) J. Biol. Chem. 274, 5995–6002.
[13] Girard, T., Cavagna, D., Padovan, E., Spagnoli, G., Urwyler, A., Zorzato, F. and Treves, S. (2001) J. Biol. Chem. 276, 48077–48082.
[14] Datar, S., Tunwell, R.E., Lehane, M., McCarthy, T.V. and Mackrill, J.J. (2003) Biochem. Trans. Soc. Meeting on Calcium Oscillations, meeting abstract (in press).
[15] Grynkiewicz, G., Poenie, M. and Tsien, R.Y. (1985) J. Biol. Chem. 260, 3440–3450.
[16] Hosoi, E., Nishizaki, C., Gallagher, K.L., Wyre, H.W., Matsuo, Y. and Sei, Y. (2001) J. Immunol. 167, 4887–4894.
[17] Graeff, R.M., Mehta, K. and Lee, H.C. (1994) Biochem. Biophys. Res. Commun. 205, 722–727.
[18] Rechsteiner, M. and Rogers, S.W. (1996) Trends Biochem. Sci. 21, 267–271.
[19] Bradford, M.M. (1976) Anal. Biochem. 72, 248–254.
[20] Funaro, A., Reinis, M., Trubiani, O., Santi, S., Di Primio, R. and Malavasi, F. (1998) J. Immunol. 160, 2238–2247.
[21] Bokkala, S. and Joseph, S.K. (1997) J. Biol. Chem. 272, 12454–12461.
[22] Zhu, C.C. and Wojcikiewicz, R.J. (2000) Biochem. J. 348, 551–556.
[23] Mackrill, J.J. (1998) Biochem. Biophys. Res. Commun. 245, 428–429.
[24] Colombo, M.I., Beron, W. and Stahl, P.D. (1997) J. Biol. Chem. 272, 7707–7712.
[25] Sullivan, K.M., Lin, D.D., Agnew, W. and Wilson, K.L. (1993) Cell 73, 1411–1422.
[26] Chou, C.L., Yip, K.P., Michea, L., Kador, K., Ferraris, J.D., Wade, J.B. and Knepper, M.A. (2000) J. Biol. Chem. 275, 36839–36846.
[27] Burlando, B., Marchi, B., Panfoli, I. and Viarengo, A. (2002) Am. J. Physiol. Cell Physiol. 283, C1461–C1468.
[28] Xue, H.H., Zhao, D.M., Suda, T., Uchida, C., Oda, T., Chida, K., Ichiyama, A. and Nakamura, H. (2000) J. Biochem. (Tokyo) 128, 329–336.