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COMMENTARY

MECHANISM OF INVERSE REGULATION OF ALPHA$_1$- AND BETA-ADRENERGIC RECEPTORS

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Receptor-mediated cellular processes utilize two distinct biochemical pathways. Certain receptor systems are coupled to the enzyme adenylate cyclase, whereas other receptors function independently of cyclic AMP (cAMP), through a calcium-sensitive mechanism triggered by an increase in polyphosphoinositide breakdown. Hormones and neurotransmitters usually act on more than one type of receptor, with both cAMP and calcium-linked receptors being available for interaction with a given endogenous regulator. For instance, certain biological effects of catecholamines are associated with stimulation or inhibition of adenylate cyclase via β- or α_2-receptors, respectively, whereas other effects are independent of cAMP and involve activation of a calcium-sensitive pathway via α_1-receptors. It has been customary to regard cAMP and calcium-linked receptors in general, and α_1- and β-receptors in particular, as distinct in organization and functioning independently of each other. However, evidence has accumulated over the years to suggest that the two systems may be functionally coupled under certain conditions [1]. Such observations have gained new importance in the light of more recent findings which indicate that cAMP and calcium-linked receptor systems may actually share some components. Receptors linked to adenylate cyclase communicate with a pair of homologous, guanyl nucleotide binding proteins that mediate stimulation (N_s protein) or inhibition (N_i protein) of adenylate cyclase respectively [2]. Recent evidence indicates that guanyl nucleotide binding proteins are also involved in hormone-stimulated calcium gating [3] and in modulating the agonist affinity of calcium-linked receptors [4]. Thus, N proteins may represent a link at the molecular level between receptors utilizing cAMP or calcium-linked pathways.

Experimental models of inverse regulation of α_1- and β-receptors

The first evidence suggesting a functional coupling between α- and β-adrenergic receptors was the observation of a temperature-dependent shift from α- to β-adrenergic responses in isolated frog hearts [5]. Although similar changes have not been detected in some of the follow-up studies (see Refs. 1 and 6), recent observations confirm the existence of temperature-dependent inverse changes in α- and β-adrenergic responses not only in myocardium [7] but also in other tissues [8, 9]. More importantly, factors other than temperature have been identified that induce an apparent "conversion" of the adrenergic receptor response [1, 10]. From such findings, two important features of the "conversion" phenomenon have emerged. (a) Different stimuli can trigger the same change in the adrenergic receptor phenotype in a given tissue. This could suggest a common underlying mechanism and is best exemplified by studies in the rat liver (see below). (b) The same stimulus can elicit directionally opposite changes in the relative dominance of α- and β-adrenergic responses in different tissues of the same animal. An example of this latter point is the effect of hypothyroidism on adrenergic receptor responses in the rat: in the heart, stimulation of the force and rate of contraction shift from a β- to an α-receptor type response [11, 12], whereas phosphorylase activation in the liver changes from an α- to a β-receptor mediated event [10, 13]. Clearly, a mechanism underlying these changes should also account for the tissue-specific differences.

Recent studies of the adrenergic regulation of liver carbohydrate metabolism have been particularly useful in providing insight into the molecular mechanism of the adrenoceptor "conversion" phenomenon. Catecholamines have important metabolic effects in the liver: they stimulate glycogenolysis and gluconeogenesis and inhibit glycogen synthesis, to name a few. Studies in the rat liver point to the unusual plasticity of the adrenergic receptor mechanism of the tissue. While in normal, adult, male rats the glycogenolytic effect of catecholamines is mediated by a typical, cAMP-independent, α_1-adrenergic receptor, the same response is mediated predominantly by β-receptors in fetal [14] or female [15] rats and in male rats under different conditions. These conditions include thyroid [13, 16] and glucocorticoid [17] deficiencies, malignant transformation [18], liver regeneration [19, 20], cholestasis [19] and fasting [21]. In addition to these *in vivo* stimuli, a conversion from α_1- to β-receptor mediated glycogenolysis has been shown to occur in primary culture [22] or short term *in vitro* incubation of isolated rat liver cells [23]. A common denominator amongst most of these conditions is the lower level of cellular differentiation. This could suggest that a

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change in the adrenergic receptor phenotype is part of the process of growth and differentiation, and that a common cellular mechanism may underlie the conversion of the receptor response triggered by different factors. The experimental model most attractive for further studies is the isolated hepatocyte in primary culture, as the change in receptor response develops relatively rapidly (4–6 hr) and under *in vitro* conditions.

Adrenergic receptors and the altered glycogenolytic response

In many of the experimental models listed above, there is evidence that the altered glycogenolytic response is selective for adrenergic agents, and affects both the α_1- and β-receptor systems. In contrast to the emerging β-adrenergic response to isoproterenol, the glycogenolytic effect of the cAMP-linked hormone, glucagon, is unchanged [16] or reduced [17, 22, 23]. Similarly, whereas α_1-adrenergic activation of phosphorylase is diminished after adrenalectomy [17], thyroidectomy [13, 24, 25] or *in vitro* incubation of hepatocytes [23], the same conditions do not affect the response to the Ca^{2+}-linked activator, vasopressin [17, 23–25]. A report of unchanged α-adrenergic and reduced vasopressin and angiotensin-induced glucose release in hepatocytes from hypothyroid rats [26] is difficult to interpret in terms of receptor-specific changes. Glucose output is the net result of glucose production and utilization, and it is not a reliable indicator of receptor interactions for hormones that can influence both processes. Furthermore, exposure of hepatocytes to α_1-agonists leads to a rapid (< 1 min) loss of the high agonist affinity form of the receptor [27], which may explain the rapid decline of phosphorylase *a* activity despite continued presence of the agonist. Since glucose output in the above study was measured over a period of 1 hr [26], marked changes in the receptor-mediated initial phosphorylase response may go unnoticed.

The inverse changes in adrenoceptor responses are evident when β-adrenergic activation of adenylate cyclase [16–18, 22] or α-adrenergic effects on calcium movements are measured [17, 24, 25], suggesting that the underlying mechanism must be at or near the receptor for both responses. Indeed, numerous studies have documented inverse changes in the densities of α_1- and β-receptors, which correspond in direction to the altered receptor response in many of the conditions listed above [4, 11, 19, 21, 28–30]. There is reason to believe, however, that these changes are not the cause of the conversion of the receptor response, and may be secondary to earlier changes in the coupling of receptors to their post-receptor pathways. Rat liver cells cultured in the presence of serum-containing media display an increase in the density of β- [29–32] and a parallel decrease in the density of α_1-receptors [29, 30]. However, these changes take 1–3 days to develop fully [29, 30], whereas the conversion of the receptor response in cells cultured under identical conditions is maximal within 8 hr [22]. Moreover, when cells are maintained in suspension in serum-free Krebs buffer, conversion of the receptor response is complete within 4 hr and is not associated with any change in receptor densities [23], thus dissociating the two events. It is possible that serum has a permissive effect on secondary changes in receptor density, as also suggested by a recent study [33]. It is more likely, however, that additional factors, such as plating of the cells, are necessary for changes in receptor numbers to manifest, as liver cells suspended in medium 199 containing 10% fetal calf serum behave the same way as cells suspended in serum-free Krebs buffer (Ishac and Kunos, unpublished). What are, then, the primary, receptor-related changes? Analysis of the displacement of radiolabeled antagonists from receptors by agonists can provide information on the level of coupling of receptors to post-receptor pathways. Shallow displacement curves have been interpreted to indicate the presence of high and low agonist affinity states. By converting all receptors into the low agonist affinity form, guanyl nucleotides can right shift and increase the slope of agonist displacement curves not only for β- but also for α_1-receptors [4, 34]. We studied the effects of prolonged (4 hr) *in vitro* incubation of isolated rat liver cells on the ability of agonists to displace radiolabeled antagonist from both α_1- and β-receptors. Hepatocytes were subjected to one cycle of rapid freezing and thawing in order to permeabilize the membrane to the guanyl nucleotide analog, Gpp(NH)p. The results of the ligand binding assays on the intact, permeabilized cells are illustrated in Fig. 1. In freshly isolated cells (0 hr), displacement of [^{3}H]prazosin by epinephrine produced shallow binding isotherms. Gpp(NH)p, 200 μM, caused a small but significant right shift of the curve which also became steeper. This is in agreement with observations in purified liver plasma membranes [4, 32], and confirms the guanyl nucleotide sensitivity of hepatic α_1-receptors. In the same cells, isoproterenol displacement of [^{125}I]pindolol binding to β-receptors was only slightly affected by Gpp(NH)p, suggesting that the lack of significant β-adrenergic effects in these cells is probably related to a low level of coupling of β-receptors to adenylate cyclase. In the "4 hr" cells, the displacement curve for α_1-receptors was steeper than in freshly isolated cells and, if anything, was slightly left-shifted by Gpp(NH)p, whereas the guanyl nucleotide effect on β-receptors was greater than in 0 hr cells. These findings could be interpreted to indicate that prolonged *in vitro* incubation of isolated liver cells leads to the uncoupling of α_1-receptors and to a simultaneous increase in coupling of β-receptors. These changes could be causally related to the parallel conversion of the adrenergic receptor response, and are similar to those observed after adrenalectomy [4].

The conclusion that α_1- and β-receptors are functionally linked at the levels of their coupling rather than being interconvertible proteins, as originally suggested [5], is more compatible with growing evidence that the ligand binding units are distinct molecules. The mammalian β_2-receptor is a 64 kD glycoprotein containing a 46 kD polypeptide of recently resolved sequence [35], while the molecular mass of the α_1-receptor binding subunit is 80 kD, as first documented in our laboratory [36]. Nevertheless, the reciprocal nature of changes in α_1- and β-receptor

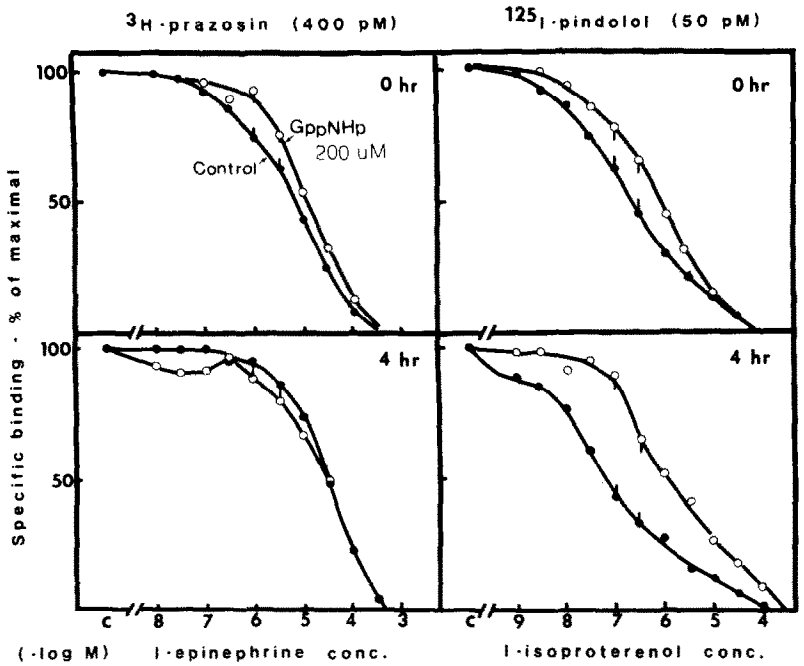

Fig. 1. Time-dependent changes in guanyl nucleotide effects on agonist affinities of α_1- and β-receptors in isolated rat liver cells. Liver cells were isolated and incubated for 30 min (0 hr) or an additional 4 hr (4 hr) in gelatin-containing Krebs buffer, as described [13]. For binding assays, cells were resuspended in 100 mM Tris–HCl + 10 mM $MgCl_2$ at pH 7.4, and subjected to one cycle of freeze-thawing at acetone/dry ice temperatures. Cells (1.5×10^6/assay tube) were preincubated for 20 min with (solid circles), or without (open circles) 200 μM Gpp(NH)p at 37° and were incubated for a further 30 min with the radiolabeled antagonist (400 pM [^{3}H]prazosin or 50 pM [^{125}I]pindolol) in the absence or presence of different concentrations of the agonist. Incubation was terminated by vacuum filtration. Points represent the mean from three (prazosin) or five (pindolol) separate experiments. Vertical bars illustrate SE for representative points.

function in a variety of conditions suggests a common regulatory factor or a common component in the larger organizational units including transducing systems. Some possibilities are discussed below.

Guanyl nucleotide regulatory proteins and adrenoceptor conversion

As pointed out above, guanyl nucleotide regulatory proteins have been implicated in the coupling of not only β- but also α_1-receptors. Recent experiments by Itoh *et al.* [37] implicate the inhibitory nucleotide binding protein, N_i, in the culture-induced conversion from α_1- to β_2-adrenergic glycogenolysis in rat liver cells. They report that primary culturing of the cells results in a gradual loss of the cellular substrate for pertussis toxin-induced ADP ribosylation. Furthermore, substances that can inhibit endogenous ADP ribosyltransferases, such as nicotinamide, are found to delay both this loss and the parallel conversion of the adrenoceptor response [37]. Itoh *et al.* propose that N_i is coupled to β_2-receptors so as to inhibit adenylate cyclase activation, and that the increase in β_2-receptor activity during primary culturing is due to the suppression of this function of N_i by the increased activity of an endogenous ADP ribosyltransferase. They further propose that the decrease in α_1-receptor function is secondary to the increase in β-receptor activity, and is thus also linked to N_i [37]. This latter part of the hypothesis, however, is not supported by more recent observations. Complete inactivation of hepatic N_i by *in vivo* treatment of rats with large doses of pertussis toxin had no effect on α_1-adrenergic activation of phosphorylase [38] or polyphosphoinositide breakdown [39]. Cholera toxin was found to be similarly ineffective [39], suggesting that guanyl nucleotide effects on hepatic α_1-receptors reflect the involvement of a novel guanyl nucleotide binding protein, tentatively labeled N_x. It is possible that the emergence of β-adrenergic functions is related to inactivation of N_i, while the parallel loss of α_1-receptor activity is due to a loss of N_x function. However, implicit in such a hypothesis is the selectivity of the interaction of N proteins with adrenergic as opposed to other receptors. Such selectivity may imply additional, receptor-specific events, such as covalent modification of the receptor proteins.

Membrane phospholipid metabolism and adrenoceptor conversion

One of the conditions associated with a conversion of the glycogenolytic response from α_1- to β-type is glucocorticoid deficiency, the effects of which can be reversed by glucocorticoid treatment [17]. In many different cell types, glucocorticoids induce the

synthesis and release of a protein which inhibits phospholipases, predominantly phospholipase A_2 [40, 41]. The structure of this protein, called lipomodulin or lipocortin, has been elucidated recently [42]. Inhibition of membrane phospholipase A_2 and the resulting decrease in the release of arachidonic acid and its eicosanoid metabolites are believed to be responsible for most of the biological effects of glucocorticoids, including those on cellular differentiation [23, 41]. The similarity between the altered adrenergic receptor response of hepatocytes caused by glucocorticoid deficiency or by primary culturing suggested to us that increased membrane phospholipase A_2 activity might be involved in both. This possibility is supported by the observation that partially purified lipomodulin acutely reverses the time-dependent conversion from α_1- to β-adrenergic glycogenolysis in isolated rat liver cells [23]. Melittin, an activator of phospholipase A_2, has opposite effects, as it rapidly induces a shift from α_1- to β-receptor responses in freshly isolated cells [23]. More recent observations indicate that increased release of arachidonic acid and subsequent formation of a cyclooxygense metabolite play a key role in this phospholipase effect. The time-dependent conversion of α_1- to β-adrenergic glycogenolysis could be prevented, if the incubation medium for the isolated hepatocytes contained fatty acid-free, but not regular, bovine serum albumin [43]. Fatty acid-free BSA avidly binds free fatty acids, and the most likely explanation of its action is removal of a fatty acid generated by the cells during prolonged *in vitro* incubation. The predominant fatty acid in the sn-2 position of membrane phospholipids is arachidonic acid. Its role in the conversion of the adrenergic receptor response is suggested by the finding that the addition of 10 μg/ml arachidonic acid to freshly isolated hepatocytes suppressed the α_1- and enhanced the β-receptor mediated activation of phosphorylase. Similar effects were not observed with stearic or oleic acids, and the effects of arachidonic acid were blocked completely by 0.2 μM ibuprofen, a cyclooxygenase inhibitor, but not by the same concentration of nordihydroguaiaretic acid, a lipoxygenase inhibitor [43]. The time-dependent shift from α_1- to β-adrenergic glycogenolysis was also attenuated by ibuprofen or indomethacin, although the concentrations of the antagonists required to inhibit these endogenously triggered changes were somewhat higher than the concentrations that block the effect of exogenous arachidonic acid. Also, repeated addition of the antagonists is necessary in experiments involving prolonged incubation of hepatocytes, to offset the loss of drug due to rapid uptake and metabolism by the cells [10]. The above findings strongly suggest that an arachidonate metabolite possibly generated through the cyclooxygenase pathway is involved in the inverse regulation of α_1- and β-receptors in the rat liver [43]. Although the possible mechanism of this regulatory function is not known, it may involve an effect of the metabolite on the functional state of a relevant N protein (see above). Experiments are in progress in our laboratory to identify this arachidonate metabolite and its possible mechanism of action on adrenoceptors in rat hepatocytes.

Protein factor(s) and conversion of the adrenoceptor response

Inhibition of protein or mRNA synthesis by cycloheximide or actinomycin D, respectively, prevents the conversion of α_1- to β-adrenergic glycogenolysis [22, 30–32, 43, 44] as well as the reciprocal changes in α_1- and β-receptor densities in primary cultured hepatocytes [30]. It is unlikely that the protein involved is the receptor protein itself, for two reasons: (1) the inhibitors prevent not only the increase in the density and reactivity of β-adrenergic receptors, but also the opposite changes in the α_1-receptor system; (2) cycloheximide and actinomycin D prevent the conversion of the adrenoceptor response during short-term *in vitro* incubation of isolated hepatocytes when there is no associated change in receptor numbers [43, 44]. This suggests the existence of a protein factor involved in the inverse regulation of α_1- and β-receptors.

Culturing of cells and the associated loss of differentiated functions is known to be accompanied by a shift from proteolytic to protein synthetic activities. One could hypothesize that, in the well-differentiated, *in situ* liver cell, the level of the protein factor involved in the conversion of the adrenoceptor response is kept low by its continuous proteolytic degradation but is increased during primary culturing due to a decline in proteolytic activity. Indeed, we have reported that the response pattern of freshly isolated rat liver cells can be acutely shifted from an α_1- to a mixed α-β type by a short exposure to phenylmethylsulfonylfluoride (PMSF), a serine protease inhibitor [44]. PMSF also mimicks the effects of prolonged incubation on the glycogenolytic response to glucagon, which is reduced, and the effect of vasopressin, which remains unchanged [44]. These findings suggest the involvement of a serin protease in the degradation of a "conversion" factor. In this context, it is noteworthy that a seryl trypsin-like protease has been isolated from plasma membrane of differentiated liver cells [45], and it has been proposed to control the levels of cell surface proteins that regulate cell growth and hepatocyte-specific functions [46]. The mechanism by which a protein factor could inversely regulate the level of coupling of α_1- and β-receptors is not known. In view of the foregoing discussion, activation of membrane phospholipase A_2, inhibition of lipocortin activity, or regulation of N protein function through activation of an endogenous ADP ribosyl transferase are possibilities to explore.

Possible role of protein kinase C

The calcium/phospholipid dependent protein kinase (protein kinase C) has a pivotal role in cellular signal transduction and cell proliferation [47]. Hormonal mediators that act through the breakdown of inositol phospholipids generate two substances with second messenger function: inositol trisphosphate (IP_3) and diacylglycerol. In rat liver, α_1-adrenergic agonists stimulate IP_3 production, which leads to the release of intracellular calcium and the subsequent activation of the glycogenolytic cascade. Calcium-linked hormones including α_1-adrenergic agonists also generate diacylglycerol, which activates protein

Table 1. Cytosol to membrane translocation of protein kinase C induced by phorbol ester (PMA) or by prolonged *in vitro* incubation of isolated rat liver cells

	^{32}P Incorporated (pmoles/mg protein/min)		
	0-hr Cells	0-hr Cells, PMA	4-hr Cells
Soluble	255 ± 46	83 ± 21*	86 ± 28*
Particulate	37 ± 16	115 ± 27*	121 ± 43*

Protein kinase C activity in cytosol and membrane fractions of hepatocytes was determined by measuring the transfer of ^{32}P from [^{32}P]-ATP to histone, and subtracting from the transfer in the presence of added phospholipids the transfer observed in their absence, as described [54]. Values are mean ± SE, N = 10. Exposure of 0-hr cells to 100 ng/ml PMA was for 10 min.

* Significant difference from corresponding value in 0-hr control cells ($P < 0.01$).

kinase C. In certain cells, calcium mobilization and protein kinase C activation are synergistic in eliciting the full biological response to such hormonal stimuli [47]. However, the relationship between these two signals appears to be more complex in the rat liver. While protein kinase C activation may be involved in α_1-adrenergic effects on cellular growth and proliferation [48], it does not contribute to α_1-receptor mediated glycogenolysis. In fact, activation of protein kinase C by phorbol esters inhibits the glycogenolytic response of isolated rat liver cells to α_1-receptor agonists, but not to vasopressin [49–52]. This selective inhibition of the α_1-adrenergic response, which has been attributed to phosphorylation of the α_1-receptor protein [51], is remarkably similar to the effects of primary culturing on hepatocyte responses. This and the known effects of phorbol esters in inhibiting differentiation and promoting growth in various cell types [53] could suggest that activation of protein kinase C is involved in the culture-induced changes in adrenoceptor responses in rat hepatocytes. Preliminary observations in our laboratory support this possibility. As shown by the data in Table 1, a 20-min exposure of freshly isolated hepatocytes to 100 ng/ml of phorbol 12-myristate,13-acetate (PMA) caused translocation of protein kinase C activity from the cytosol to the plasma membrane. A similar translocation of protein kinase C activity was observed in cells incubated for 4 hr in the absence of PMA. Thus, prolonged *in vitro* incubation of cells is associated with activation of protein kinase C by an endogenous process. We also tested the effects of PMA on the hormone-induced activation of glycogen phosphorylase in freshly isolated rat liver cells. A 2-min exposure to 2 ng/ml PMA markedly suppressed the effect of phenylephrine, moderately increased the effect of isoproterenol, and did not influence the effects of vasopressin or glucagon (Table 2). The same concentration of phorbol 12-monoacetate, an analog that does not activate protein kinase C, did not influence either the phenylephrine or the isoproterenol response of hepatocytes (Table 2). The changes caused by PMA are similar but not identical to those observed after prolonged incubation of liver cells in the absence of PMA. While the α_1-receptor-mediated response was nearly completely and consistently suppressed, the potentiation of the effect of isoproterenol was smaller after PMA than in 4-hr cells, and less consistently observed. Also, PMA does not influence isoproterenol-induced cAMP accumulation in liver cells from female rats [51]. Thus, although activation of protein kinase C may fully account for the time-dependent reduction of the α_1-receptor response, additional factors must also contribute to the emergence of a strong β-adrenergic response.

What is the relationship between the phospholipase/arachidonic acid mechanism described above and protein kinase C in the conversion of the adrenoceptor response? Activation of protein kinase C can lead to increased release and metabolism of arachidonic acid [53] and, thus, may trigger the formation of a key arachidonate metabolite involved in receptor regulation. However, preincubation of hepatocytes with 2 μM ibuprofen failed to prevent the effects of PMA on the adrenergic activation of phosphorylase (Ishac and Kunos, unpublished). Alternatively, the sequence may be the reverse whereby increased release of arachidonic acid or one of its metabolites may activate protein kinase C. Indeed, direct activation of purified protein kinase C by arachidonic acid has been demonstrated

Table 2. Effects of phorbol esters on hormone-induced glycogenolysis in freshly isolated rat hepatocytes

	^{14}C Incorporated (nmoles/mg protein/min)			
	Phenylephrine 10 μM	Isoproterenol 1 μM	Vasopressin 0.01 μM	Glucagon 0.01 μM
Control	25.7 ± 2.0	5.1 ± 1.4	32.6 ± 7.4	54.1 ± 1.5
PMA, 2 ng/ml	8.6 ± 1.3*	11.7 ± 1.6†	32.2 ± 1.5	64.1 ± 4.4
PA, 2 ng/ml	22.6 ± 1.6	5.0 ± 0.8		

Phosphorylase *a* activity was measured by the incorporation of [^{14}C]-glucose-1-P into glycogen, as described [13]. Values (mean ± SE) represent the increase in enzyme activity over basal (17.6 ± 0.7) by a maximally effective hormone concentration; N = 12 for phenylephrine and isoproterenol, and N = 3 for vasopressin and glucagon. Exposure to PMA (phorbol 12-myristate 13-acetate) or PA (phorbol 12-monoacetate) was for 4 min.

*,† Significant difference from control: *$P < 0.005$, and †$P < 0.05$.

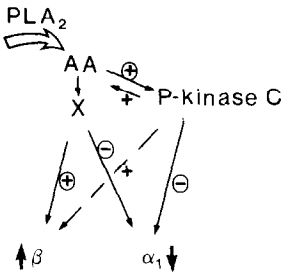

Fig. 2. Hypothesis for mechanisms of inverse regulation of α_1- and β-adrenoceptors. Abbreviations: PLA_2, phospholipase A_2; AA, arachidonic acid; X, cyclooxygenase metabolite of AA; (+) and (−) indicate stimulation and inhibition in the indicated direction respectively.

[55], and in preliminary experiments we found cytosol to membrane translocation of protein kinase C by exposure of isolated hepatocytes to 10 μg/ml arachidonic acid. It is also possible that arachidonic acid and protein kinase C are involved in parallel, synergistic regulatory mechanisms, as it has been shown to be the case in certain other systems [56]. We tentatively suggest the scheme illustrated in Fig. 2, although much more work is required to substantiate various aspects of this regulatory system.

Potential implications of the inverse regulation of α_1- and β-adrenoceptors

The findings described in the foregoing sections were in so-called unidirectional systems, where stimulation of α_1- and β-receptors results in a similar biological end response, i.e. increased rate and force of myocardial contractility or increased hepatic glycogenolysis. In such systems, the significance of a change in the relative contribution of the two receptors to the net response may not be obvious. Altered sensitivity to endogenous catecholamines [11] or changes in the energy requirement for a given tissue response may be the consequence of a "conversion", although definitive proof is lacking. Potentially more important is the realization that catecholamines cause not only acute effects but can also influence growth and differentiation, and a change in receptor type may influence these functions. The role of hepatic α_1-receptors in stimulating DNA synthesis and growth [48] and the conversion from α_1- to β-type responses in conditions associated with a lower level of differentiation of the liver may reflect the interplay of such regulatory influences.

The possible functional significance of inverse changes in α_1- and β-receptor activity should be more obvious in bidirectional systems, where the two receptors mediate opposite effects. An autonomic imbalance including hypoactivity of β- and hyperactivity of α-receptors in lung tissue has been documented in bronchial asthma (see Ref. 57), and there are examples of similar changes in cystic fibrosis [58] or in certain tissues of animals with experimental hypertension [59]. The molecular mechanisms discussed in this review should serve to stimulate inquiries about the cellular defects underlying the altered autonomic reactivity in certain human disease conditions.

Note added in proof: A paper published after this review had been submitted [60] indicates that exposure of rats to constant light increases β and decreases α_1-adrenoceptor responses of isolated pinealocytes, and that these changes can be mimicked by exposure of pinealocytes from control rats to mepacrine, a phospholipase A_2 inhibitor. This confirms the role of phospholipase A_2 in the inverse regulation of α_1 and β-receptors, although the direction of the change in the pineal is opposite to that in the liver.

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