Interleukin-8 Synthesis, Regulation, and Steroidogenic Role in H295R Human Adrenocortical Cells


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The adrenal gland secretes several cytokines, and cytokines modulate steroid secretory function by this gland. In this study, a survey of cytokine production by H295R human adrenocortical cells demonstrated that these cells secreted IL-2, IL-4, IL-8, IL-10, IL-13, and TNFα but not IL-5, IL-12, or interferon-γ. IL-8 was the IL secreted at higher concentration. IL-8 secretion, its regulation, and role in steroidogenesis were further studied. Secreted ILs and steroids were measured by ELISA in cell culture supernatant. IL-8 mRNA was quantified by real-time RT-PCR. H295R cells and human adrenal gland expressed IL-8 mRNA. Angiotensin II, potassium, endothelin-1, IL-1α, IL-1β, TNFα, and Escherichia coli lipopolysaccharide dose-dependently increase IL-8 secretion by H295R cells after 24 h incubation. IL-6 had no effect on IL-8 secretion. Angiotensin II time-dependently increased IL-8 secretion by H295R cells up to 48 h. Angiotensin II caused a biphasic increase in IL-8 mRNA expression with a peak 6 h after stimulation. TNFα synergized angiotensin II, potassium, and IL-1α-mediated IL-8 secretion. IL-8 did not modify aldosterone or cortisol secretion by H295R cells under basal or stimulated (angiotensin II or potassium) conditions. In conclusion, it is demonstrated for the first time that human adrenal cells expressed and secreted IL-8 under the regulation of angiotensin II, potassium, endothelin-1, and immune peptides. Adrenal-secreted IL-8 is one point of convergence between the adrenal gland and the immune system and may have relevance in physiological and pathophysiological conditions associated with increased levels of aldosterone secretagogues and the immune system. (Endocrinology 147: 891–898, 2006)
cells, neutrophils, natural killer cells) and nonimmune cells (endothelial cells, fibroblasts, epithelial cells, adipocytes). IL-8 is not constitutively produced by most cell types but can be rapidly induced by several factors, including ILs (IL-1, TNFα), bacteria, and viruses (27, 28). IL-8 expression is regulated at the transcription level by coordinated stimulation of nuclear factor-κB and JUN-N-terminal protein kinase pathways (27, 28). IL-8 mRNA stability is enhanced by upregulation of the p38 MAPK. IL-8 binds two distinct types of receptors, CXCR1 and CXCR2, with similar high affinity. The main biological activity of IL-8 is as a neutrophil chemotactic factor. IL-8 has several other effects on neutrophils, including degranulation, increasing intracellular calcium concentration, respiratory burst, and promotion of adherence to endothelial cells. IL-8 also regulates several biological activities in basophils, eosinophils, T cells, and B cells, including migration, chemotaxis, and intracellular calcium concentrations (25, 26). IL-8 regulates several cellular and tissue activities in nonimmune cells. IL-8 promotes angiogenesis by inducing migration and proliferation of endothelial cells (29). IL-8 is chemotactic and mitogenic for keratinocytes (30). IL-8’s role in pathology includes not only the well-known recruitment of leukocytes to infection zones but also several others. IL-8 participates in reperfusion injury after ischemia, and this damage can be blocked with anti-IL-8 antibodies. IL-8 is increased in synovial fluid of patients with rheumatoid arthritis (31) and is involved in neovascularization, which is required for panus formation (32).

We screened H295R human adrenocortical cells for cytokine production and its regulation by the aldosterone secretagogues angiotensin II and potassium. Of several cytokines studied [IL-2, IL-4, IL-5, IL-8, IL-10, IL-12, IL-13, interferon (IFN)-γ and TNFα], we found that IL-8 was the IL with the highest secretion levels and that it was increased by angiotensin II and potassium. Of several cytokines studied [IL-2, IL-4, IL-5, IL-8, IL-10, IL-12, IL-13, interferon (IFN)-γ and TNFα], we found that IL-8 was the IL with the highest secretion levels and that it was increased by angiotensin II and potassium. The present study focus on the regulation of IL-8 secretion in human adrenal cells and its role on steroidogenesis.

Materials and Methods

Cell culture

H295R human adrenocortical cells (a generous gift from Dr. W. E. Rainey, University of Texas Southwestern, Dallas, TX) were grown in complete media containing DMEM-F12 (1:1) supplemented with 2% Ultroser G (Biosera, Villeneuve-la-Garenne, France), ITS-Plus (Discovery Labware, Bedford, MA), and antibiotic/antimycotic mixture (In- vitrogen, Carlsbad, CA). Cells were grown until approximately 95% confluent in 6-well plates. Media were replaced with 2 ml fresh media vitrogen, Carlsbad, CA). Cells were grown until approximately 95% confluent in 6-well plates. Media were replaced with 2 ml fresh media. Cells were grown until approximately 95% confluent in 6-well plates. Media were replaced with 2 ml fresh media.

Cytokine ELISA

Secreted cytokines (IL-2, IL-4, IL-5, IL-8, IL-10, IL-12, IL-13, IFNγ, and TNFα) were quantified in cell culture supernatants using the commercial multiplex sandwich ELISA kit SearchLight human TH1/TH2 cytokine array 1 (Endogen, Rockford, IL). Incubations were performed in duplicates and IL secretion quantified by serial dilutions in duplicates. Media incubated without cells were used as background and subtracted to generate reported values. Assay sensitivity, as reported by the manufacturer, was 0.2 pg/ml for IL-2, IL-5, IL-10, and IFNγ; 0.4 pg/ml for IL-8, IL-12, and TNFα; and 0.8 pg/ml for IL-13 and TNFα.

Secreted IL-8 was also determined in cell culture supernatants by sandwich ELISA using the OptEIA human IL-8 set (PharMingen, San Diego, CA). Assays were performed following the manufacturer’s instructions except for the following modifications. Avidin-horseradish peroxidase conjugate was replaced with Zymad streptavidin-horseradish peroxidase (Zymed, San Francisco, CA) at a final concentration of 1.25 µg/ml. Plates were developed with 200 µl/well of sodium citrate buffer (pH 5.0) containing 0.1 mg/ml tetramethylbenzidine and 0.05 mg/ml/mole urea peroxidase for 1 h at room temperature. The reaction was stopped with 100 µl/well of 1 N H2SO4, and read at 450 nm and the standard curve adjusted to a four-parameter equation. Assay sensitivity was 6.25 pg/ml for recombinant human IL-8.

Aldosterone ELISA

Aldosterone was measured in cell culture supernatant by ELISA using a monoclonal antibody as previously described (33). Assay sensitivity was 20 pg/ml for aldosterone.

Cortisol ELISA

Cortisol was measured in cell culture supernatant by ELISA using a polyclonal antibody previously described (34) with the following modifications. Briefly, high-binding 96-well plates (Griener, catalog no. 655061) were coated with anticortisol antibody (1:3500, 100 µl/well) in 0.1 M borate buffer (pH 9.0) overnight at room temperature. Plates were washed four times with washing buffer (PBS with 0.05% Tween 20). Triplicate 10-µl samples or standards were incubated with 16 ng/ml cortisol-peroxidase conjugate (Sigma) in 300 µl assay buffer (PBS with 0.066% sodium casein and 0.05% Tween 20) per well and incubated overnight at room temperature. Plates were washed four times with washing buffer and developed as described for IL-8 ELISA. Assay sensitivity was 1 ng/ml for cortisol.

Real time RT-PCR

Human adrenal total RNA was obtained from several sources: hAd1 (59 yr old male donor) from Biochain Institute, Inc. (Hayward, CA), hAd2 (pooled from 61 male/female Caucasian donors, aged 15–61 yr) from BD Biosciences (Mountain View, CA), and hAd3 (30-yr-old female donor) from Stratagene (La Jolla, CA). H295R cell total RNA was extracted with Tri-Reagent (MRC, Cincinnati, OH), resuspended in diethyl pyrocarbonate-H2O, DNase treated with DNA-free kit (Ambion, Austin, TX), and quantified by spectrophotometry. Five micrograms of RNA were reverse transcribed (RT) with 0.5 µg of T, VN primer and SuperScript II (Invitrogen) in a final volume of 20 µl. The reaction was carried out for 50 min at 42 C and terminated by incubation at 70 C for 15 min.

Real-time PCR was performed with 1 µl RT product, 1 µl titanium Tag DNA polymerase (CLONTECH, Palo Alto, CA), 1:20,000 dilution SYBR Green I (Molecular Probes, Eugene, OR), 0.2 mM deoxynucleotide triphosphates, and 0.1 µM of each primer. Cycling conditions were 1 min at 95 C, 50 cycles of 15 sec at 95 C, 15 sec at 60 C, and 1 min at 72 C. Primer for IL-8 sense 5’-AGGTGCAAGTTTGCACAGA-3’ and antisense 5’-TTTCTTGTTGCGCAGTGTT-3’ (www.realtimeprimers.org). Glyceraldehyde-3-phosphate dehydrogenase (GAPDH) primers were already described (35). Real-time data were obtained during the extension phase, and critical threshold cycle values were obtained on the log phase of each gene amplification curve. PCR product quantification was performed by the relative quantification method (36). Efficiency for each primer pair was assessed by using serial dilutions of RT product. Results are expressed as arbitrary units and normalized against GAPDH mRNA expression.

Statistics

All results are expressed as mean ± SEM. Results were analyzed by ANOVA followed by Dunnett (comparisons vs. control) or Tukey (multiple comparisons) contrasts using the Graphpad Prism package (version...
4.02; Graphpad Software, Inc., San Diego, CA). TNFα potentiation was defined as the fold induction of \((\text{inducer/TNF} \alpha /\text{H9251 baseline})/(\text{inducer + basal/TNF} \alpha - 2 \times \text{basal})\).

Results

**H295R cells secrete several cytokines**

H295R human adrenocortical cells were screened for several cytokine secretions. H295R cells were incubated with increasing concentrations of angiotensin II, potassium, ET-1, forskolin, and PMA for 24 h and ILs measured in the cell culture supernatant by multiplex ELISA (Fig. 1). H295R cells secrete IL-2, IL-4, IL-8, IL-10, IL-13, and TNFα but did not secrete IL-5, IL-12, or IFNγ. IL-8 was the IL secreted at higher concentration. Secretion of IL-8 by H295R cells was up-regulated by angiotensin II, potassium, ET-1, and PMA. Angiotensin II increased IL-10 and IL-13 secretion but decreased IL-2 and TNFα secretion. Potassium increased IL-2 and IL-10 secretion. ET-1 increased IL-2, IL-10, and IL-13 secretion but decreased TNFα secretion by the H295R cells. Forskolin is an activator of adenylate cyclase that increases intracellular cAMP and elicits cAMP-dependent cellular responses mainly through protein kinase A activation. Forskolin decreased IL-4 and TNFα secretion by H294R cells after a 24-h incubation. PMA, an activator of protein kinase C, increased IL-2, IL-4, IL-10, and IL-13 secretion but decreased TNFα secretion. We focused our studies on IL-8 because the rate of its secretion was the highest of the cytokines measured and its secretion was positively regulated by the aldosterone secretagogues angiotensin II, potassium, and ET-1 in H295R cells.

**Adrenal gland and H295R cells express IL-8 mRNA**

Human adrenal gland and H295R human adrenocortical cells express IL-8 mRNA (Fig. 2). Figure 2 shows an agarose gel electrophoresis of IL-8 PCR products with both adrenal glands and H295R cells RNA as template. Negatives controls with no RT reaction show no amplification. GAPDH was used as housekeeping control gene.
Regulation of IL-8 secretion in H295R cells

The basal secretion of IL-8 by H295R cells was 30.0 ± 1.5 pg/ml. To assess possible modulators of IL-8 secretion, H295R cells were incubated with increasing concentrations of different agents for 24 h and IL-8 measured in the culture medium by sandwich ELISA (Fig. 3). Angiotensin II increased IL-8 secretion dose dependently, reaching a plateau at a concentration of 100 nM (570 ± 30 pg/ml), Potassium (16 and 20 mM) dose-dependently increased IL-8 secretion, reaching a maximum of 100 ± 2.1 pg/ml at a potassium concentration of 20 mM. ET-1 caused a modest increase (2.3-fold) at 10 nM, reaching a value of 69 ± 6.8 pg/ml. IL-1α and -1β were the most potent inducers of IL-8 secretion and produced a biphasic response curve with maximal stimulatory concentrations of 10 and 1 ng/ml, respectively, producing 3900 ± 490 and 4100 ± 100 pg/ml-d IL-8. TNFα increased IL-8 secretion at 10 and 100 ng/ml, reaching values of 200 ± 16 pg/ml-d at 10 ng/ml. IL-6 did not modify IL-8 secretion at concentrations between 0.1 and 100 ng/ml. E. coli LPS serotypes 0111:B4 and 055:B5 at concentrations of 100 ng/ml increased IL-8 secretion to 150 ± 7.5 and 89 ± 3.1 pg/ml-d, respectively.

Dexamethasone modulates IL-8 secretion mediated by several agents in other systems (37, 38). H295R cells were incubated with angiotensin II (100 nM), potassium (16 mM), IL-1α (10 ng/ml), IL-1β (1 ng/ml), TNFα (10 ng/ml), or E. coli LPS 0111:B4 (100 ng/ml) in the presence or absence of 10 μM dexamethasone. Dexamethasone coincubation did not modify IL-8 secretion in H295R cells mediated by any of these agents.

Angiotensin II increases IL-8 mRNA and protein in a time-dependent manner

Angiotensin II (100 nM) time-dependently up-regulated IL-8 protein secretion in H295R cells (Fig. 4). Angiotensin II treatment increased IL-8 accumulation in the culture media from 6 to 48 h. Maximal angiotensin II-mediated IL-8 accumulation was observed after 24 h incubation (1085 ± 11 vs. 3.0 ± 1.1 pg/ml-d). Angiotensin II increased IL-8 mRNA levels between 3 and 12 h, peaking at 6 h after treatment. Maximal induction (33-fold) was observed after 6 h angiotensin II treatment. After 24 h treatment, IL-8 mRNA levels returned to basal levels, similar to that of untreated cells.
Angiotensin II-mediated aldosterone and cortisol secretion increased steadily over the 48-h incubation period.

**TNFα synergy of IL-8-stimulated secretion**

TNFα and IL-1 are the strongest inducers of IL-8 secretion in most cell types. TNFα is a moderate inducer of IL-8 secretion in H295R cells by itself. To study whether TNFα modulates IL8 secretion in H295R cells mediated by aldosterone secretagogues, H295R cells were coincubated with angiotensin II (100 nM), potassium (16 mM), or IL-1α (1 ng/ml) in the presence or absence of 50 ng/ml TNFα for 24 h and IL-8 secretion determined in the cell culture supernatants. *P < 0.05, **P < 0.01, vs. time = 0.

**Protein synthesis inhibition and IL-8 secretion**

To study the requirement of newly synthesized protein in angiotensin II-mediated IL-8 secretion, H295R cells were incubated with or without angiotensin II (100 nM) in the presence or absence of 10 μg/ml cycloheximide for 24 h and IL-8 and steroid secretion quantified in cell culture supernatant (Fig. 6). As expected, cycloheximide completely blocked angiotensin II-mediated aldosterone and cortisol secretion; however, the protein synthesis inhibitor blocked only 20% of the angiotensin II-mediated IL-8 secretion.

**Cytokines and steroidogenesis**

Angiotensin II and potassium, the most potent known inducers of aldosterone synthesis, also induce IL-8 secretion in H295R cells. Therefore, we studied whether IL-8 could modulate aldosterone or cortisol production. H295R cells were incubated with increasing concentrations (0.1–100 ng/ml) of either IL-8 isoform, 72 or 77 amino acids, under basal or stimulated (10 nM angiotensin II or 12 mM potassium) for 24 h. Neither IL-8 isoform modified aldosterone or cortisol secretion under basal or stimulated (angiotensin II or potassium) conditions (data not shown).

To determine whether IL-8 is necessary for aldosterone production, H295R cells were incubated with increasing concentrations (0.05–5 μg/ml) of a neutralizing anti-IL-8 antibody under basal or stimulated (10 nM angiotensin II or 12 mM potassium) for 24 h. Neutralizing anti-IL-8 antibody did not modify aldosterone secretion under basal or stimulated (angiotensin II or potassium) conditions (data not shown).

**Fig. 4.** IL-8 mRNA expression and IL-8 and steroid secretion by angiotensin II-stimulated H295R cells. H295R cells were incubated with 100 nM angiotensin II for the time periods indicated. IL-8 mRNA was quantified by real-time RT-PCR. IL-8 protein, aldosterone, and cortisol were quantified by ELISA in cell culture supernatants.

**Fig. 5.** TNFα potentiation of IL-8 secretion. H295R cells were incubated with 100 nM angiotensin II, 16 mM potassium (K+), or 1 ng/ml IL-1α in the presence or absence of 50 ng/ml TNFα for 24 h. IL-8 secretion was quantified in cell culture supernatant by ELISA. ##P < 0.01, ###P < 0.001, TNFα vs. non-TNFα. All treatments were significant (P < 0.05) vs. basal without TNFα.
In this study we demonstrate that: 1) the human adrenal gland and human adrenocortical cells H295R secrete IL-8 and express IL-8 mRNA; 2) IL-8 secretion by H295R is increased by angiotensin II, potassium, and IL-1α on IL-8 secretion by H295R cells; and 4) IL-8 did not modify aldosterone or cortisol secretion. To the best of our knowledge, this is the first report of the modulation of IL secretion in the adrenal by potassium and ET-1. These results suggest that the adrenal gland plays a central role, connecting not only the HPA axis and the immune system but also between the immune system and regulators of salt and water homeostasis and blood pressure. Whether aldosterone secretagogues such as angiotensin II, potassium, or ET-1 become important modulators of the immune system under certain pathological conditions remains unknown.

The renin-angiotensin-aldosterone system and the immune system interact within the adrenal gland. In one hand, angiotensin II dose-dependently up-regulates IL-6 secretion by rat adrenal zona glomerulosa cells (39). On the other hand, several cytokines modulate mineralocorticoid secretion. IL-1β decreases both basal and angiotensin II-stimulated aldosterone production by rat adrenal zona glomerulosa cells in vitro, suggesting a direct effect of IL-1β on adrenal cells. IL-1β dose-dependently decreases angiotensin II-mediated aldosterone secretion, and this effect can be blocked with anti-IL-1 antibodies (22). IL-1β increases plasma renin activity, plasma aldosterone, and blood pressure in hypophysectomized rats (17). IL-6 secretion by the adrenal gland is increased by angiotensin II, and IL-6 increases aldosterone secretion by primary cultures of human adrenal cells (12). TNFα decreases angiotensin II-mediated aldosterone production by rat adrenal glomerulosa cells but does not alter basal aldosterone secretion (22). IL-6 and TNFα decrease angiotensin II-mediated aldosterone secretion by primary cultures of bovine adrenal zona glomerulosa cells, but it increases basal and ACTH-mediated aldosterone secretion (40). Our data show that although IL-8 secretion by adrenal cells is up-regulated by aldosterone secretagogues, IL-8 did not modify aldosterone or cortisol secretion under basal or angiotensin II-stimulated conditions. Furthermore, adrenal cell-secreted IL-8 seems not to be required for basal or angiotensin II-stimulated conditions for aldosterone secretion because a neutralizing antibody against IL-8 did not modify steroid secretion. Adrenal secreted IL-8 could act in paracrine or exocrine mode. These results suggest that if IL-8 acts in a paracrine mode, its physiological target is not steroidogenesis regulation.

Discussion

In this study we demonstrate that: 1) the human adrenal gland and human adrenocortical cells H295R secrete IL-8 and express IL-8 mRNA; 2) IL-8 secretion by H295R is increased by angiotensin II, potassium, and endothelin-1 as well as several inflammatory modulators including IL-1α, IL-1β, TNFα, and LPS; 3) TNFα has a synergic interaction with angiotensin, potassium, and IL-1α on IL-8 secretion by H295R cells; and 4) IL-8 did not modify aldosterone or cortisol secretion. To the best of our knowledge, this is the first report of the modulation of IL secretion in the adrenal by potassium and ET-1. These results suggest that the adrenal gland plays a central role, connecting not only the HPA axis and the immune system but also between the immune system and regulators of salt and water homeostasis and blood pressure. Whether aldosterone secretagogues such as angiotensin II, potassium, or ET-1 become important modulators of the immune system under certain pathological conditions remains unknown.

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The H295R cells used for these studies are a well-established in vitro model to study cortical adrenal gland physiology and regulation. H295R cells are pluripotent and the only available cell line that retains a normal steroid production profile, including regulation by angiotensin II and potassium similar to that of normal adrenal cells provided that the cell culture conditions are adequate. Our results differ from those reported by Schteingart et al. (41). They reported a clinical syndrome in which a patient with adrenocortical carcinoma presented with hypertension, history of arthritis, fever, leukocytosis, and increased acute phase reactants. Cells obtained from the tumor secreted IL-8 in a time-dependent manner in vitro and increased plasma IL-8 when transplanted into immunodeficient mice. However, the NCI-H295 cells, the progenitor cell line of H295R cells, were used...
as the negative nonsecreting chemokine adrenal cell line for this study. The difference between these results and ours is probably due to cell culture conditions and more sensitive analytical methods used in the present report. H295R cells are very dependent on cell culture conditions to maintain its differentiated adrenal-secreting cell phenotype, and the conditions used in the present report allow the growth of H295R cells while maintaining its mineralo- and glucocorticoid secretion pattern and regulation similar to freshly isolated human adrenal cells (42). The results presented in the present report suggest that H295R cells secrete lower levels of IL-8 than the tumor cells in the Schteingart report and that this secretion is finely regulated. Whereas cytokine production by the adrenal gland tumor was probably autonomous, interpretation of this patient report should consider our results indicating that the secretion of IL-8 by the adrenal gland is modulated by several factors.

IL-8 secretion is regulated by glucocorticoids in several systems (37, 38). Conversely, in the present study, the synthetic glucocorticoid dexamethasone did not modify IL-8 secretion by H295R cells under basal or stimulated conditions. A similar lack of regulation by dexamethasone has been observed in rat adrenal zona glomerulosa cells that secrete IL-6 under basal, IL-1β-, or ACTH-mediated conditions (13).

TNFα potently synergized the angiotensin II-, potassium-, or IL-1α-induced stimulation of IL-8 secretion by H295R cells. TNFα caused a 30-fold potentiation of angiotensin II-mediated IL-8 secretion, reaching values of approximately 4.5 nm in 24 h, a value similar to those reported in other nonimmune tissues that produce IL-8 (37). Although IL-8 secretion levels by adrenal glands in vivo are unknown, these in vitro studies suggest that they could have an important physiological role. IL-8 serum levels in healthy subjects have been determined to be in the picomolar range (43). IL-8 secretion up-regulated by aldosterone secretagogues and their potentiation by TNFα could reach significant levels to elicit biological effects. Heart failure (44–46), cardiac cachexia (47), and cirrhosis with spontaneous bacterial peritonitis (48) have been associated with abnormal elevation of serum TNFα and activation of renin-angiotensin system in human patients.

The source of IL-8 secretagogues could be the circulation or the adrenal gland itself. The adrenal gland is one of the main target organs of circulating angiotensin II. This peptide can be generated also in the adrenal gland as an intraadrenal renin-angiotensin system has been reported both in vivo and in vitro (49, 50). Adrenal cells secrete IL-1 and TNFα, both potent stimulators of IL-8 secretion by H295R cells. IL-1 and TNFα are, along with IL-6, the three most important cytokines secreted during the immune response. These cytokines are also produced by macrophages, and macrophages have been demonstrated in all layers of the human adrenal gland, frequently attached to the endothelial wall but also in direct contact with cells of the adrenal cortex (51). Coincubation of human adrenal cells and monocytes indicates that immune cells significantly increase cortisol secretion (52). Lymphocyte infiltration into the adrenal gland is increased in both physiological and pathophysiological conditions. The infiltration is elevated in elderly subjects over the age of 70 yr, compared with adults younger than 49 yr old (53). A patient with 21-hydroxylase deficiency has been reported to present a high level of B and T lymphocyte infiltration in the adrenal gland (54). A cross-talk between the adrenal gland cells and the immune system has been suggested for its hyperandrogenism and adenocortical growth (54). All these data suggest that the adrenal gland is subjected to endogenous and circulating changing levels of cytokines.

The lack of effect of the protein synthesis inhibitor cycloheximide on angiotensin II-mediated IL-8 secretion contrasts with the almost complete blockade of angiotensin II-mediated aldosterone secretion induction. Aldosterone secretion dependence on protein synthesis is a well-known effect. The lack of effect of cycloheximide on IL-8 secretion suggests that although angiotensin II up-regulates IL-8 mRNA levels, most of the effect on IL-8 secretion is through an increase of already synthesized IL-8 stored intracellularly.

The function of adrenal-secreted IL-8 is currently unknown. An important synergic effect on IL-8 secretion was observed between aldosterone secretagogues and TNFα. Several physiological and pathophysiological conditions that develop with increases in both the renin-angiotensin system and immune system, which later on can have an impact in adrenal secreted IL-8, have been described.

In conclusion, human adrenal cells secrete and express IL-8. Its secretion is up-regulated by angiotensin II, potassium, ET-1, and immune peptides. The IL-8 paracrine or exocrine role is currently unknown because this IL did not modify aldosterone or corticosterone secretion. The fine regulation of IL-8 secretion by several agents and the synergic effect of TNFα with angiotensin II and potassium suggest that adrenal-secreted IL-8 may play a role connecting aldosterone secretagogues, the immune system, and the adrenal gland.

Acknowledgments

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The authors have nothing to declare.

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