Cocaine Causes Increased Type I Interferon Secretion by both L929 Cells and Murine Macrophages


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Cocaine has been demonstrated to have a number of different effects on immune cell functions. We have reported alterations of cellular functions by macrophages (MΦ) exposed to cocaine in vitro, including the inhibition of mouse hepatitis virus replication. Here, we present evidence that cocaine stimulates the secretion of an antiviral product that is neutralized by anti-interferon (anti-IFN). A dose-dependent increase in the secretion of IFN by both MΦ and L929 cells incubated with cocaine, with a concomitant decrease in virus replication, is also reported. The increase in IFN secretion was most pronounced when cells were cultured in the presence of the IFN inducer poly(I:C). The effect of cocaine on IFN production was found to be primarily at the transcript level in both MΦ and L929 cells. These findings further support our previous research demonstrating an antiviral activity of cocaine in vitro. The relevance of this activity to viral infections in general remains to be determined.

Cocaine has been reported to exhibit a variety of immunomodulatory effects on different immune functions (30). Previous reports by us and others have demonstrated that macrophages (MΦ) exposed to cocaine in vitro displayed increased phagocytosis of microorganisms (10), with an increased production of reactive oxygen intermediates (28, 29) as well as decreased cytokine secretion (24) and production of reactive nitrogen intermediates (27). Further studies have reported that cocaine enhanced neutrophil phagocytosis, increased natural killer (NK) cell activity and distribution (26), reduced the T-cell mitogen response (13), and reduced the cytotoxic ability of splenic immune cells (14). Taken together, these studies indicate that the effects of cocaine vary considerably depending on which parameter of the immune repertoire is investigated as well as the tools used to measure them.

The chronic use of illicit drugs has been linked to an increased susceptibility to viral infections (3, 5). In a series of studies by Peterson et al., cocaine was reported to increase the production of p24 antigen in human immunodeficiency virus (HIV)-infected, activated peripheral blood mononuclear cells (PBMC) (18–20). This increase was taken to indicate an increase in IFN production of p24 antigen in human immunodeficiency virus (HIV)-infected, activated peripheral blood mononuclear cells (PBMC) (18–20). This increase was taken to indicate an in- cluded cytokine secretion (24) and production of reactive nitrogen intermediates (27). Further studies have reported that cocaine enhanced neutrophil phagocytosis, increased natural killer (NK) cell activity and distribution (26), reduced the T-cell mitogen response (13), and reduced the cytotoxic ability of splenic immune cells (14). Taken together, these studies indicate that the effects of cocaine vary considerably depending on which parameter of the immune repertoire is investigated as well as the tools used to measure them.

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The principal nonspecific mechanism used by cells to inhibit virus replication is the stimulation of type I interferon (IFN) secretion. The two type I IFNs shown to have potent antiviral activity are IFN-α and IFN-β (IFN-α/β). These cytokines have been demonstrated to have myriad effects on cell functions, including development of an antiviral state, inhibition of cell proliferation, and enhancement of NK cell activity (22). IFN-α/β are secreted by most nucleated cells in response to viral infection and affect virus replication by inducing an antiviral state in an autocrine and paracrine fashion (see reference 2 for review). In this study, we have further characterized the effect of cocaine on virus replication in vitro. Using both primary MΦ cultures and the L929 cell line, cocaine’s effects on IFN-α/β secretion and virus replication were investigated. These studies demonstrate that cocaine’s antiviral effect is mediated through an increase in IFN secretion and that it regulates the production of IFN at least in part at the mRNA level.

MATERIALS AND METHODS

Experimental animals. Male 6- to 8-week-old C57BL/6 mice were purchased from Jackson Laboratories, Bar Harbor, Maine. All mice were housed in the Laboratory Animal Research Center at Texas Tech University Health Sciences Center and were cared for as stated in the policies and regulations of the Institutional Animal Care and Use Committee.

Culture media and reagents. Dulbecco’s modified Eagle’s medium (DMEM) (Gibco, Long Island, N.Y.) was supplemented with 2% fetal bovine serum (FBS) and 100 units/ml penicillin and 100 μg/ml streptomycin. Polyclonal anti-tumor necrosis factor alpha (anti-TNF-α) was a gift from George Gifford, Dept. of Microbiology at the University of Florida, Gainesville. Polyclonal anti-IFN-α and anti-IFN-β (10,000 U/ml) and nonimmune control rabbit serum were obtained from Access Bio- medical, San Diego, Calif. Polyclonal anti-tumor necrosis factor alpha (anti-TNF-α) was a gift from George Gifford, Dept. of Microbiology at the University of Florida, Gainesville. Polyclonal anti-IFN-α and anti-IFN-β were purchased from Sigma Chemical Co., St. Louis, Mo. Cocaine hydrochloride was obtained from the National Institute of Drug Abuse. Cocaine was dissolved in DMEM-S and filtered through a 0.22-μm filter prior to use. No cytotoxicity to cocaine was noted at any concentration employed in this study, as indicated by trypan blue exclusion. All media and reagents were tested for lipopolysaccharide (LPS) using the Limulus amoebocyte lysate assay (Associates of Cape Cod, Cape Cod, Mass.) and were found to contain less than 0.1 ng of LPS per ml.

MΦ collection and preparation. To generate inflammatory peritoneal MΦ, mice were injected intraperitoneally with 1 ml of 3% thioglycollate broth (Baltimore Biological Laboratories, Baltimore, Md.). MΦ were collected 4 days later and cultured as described previously (9). Briefly, a peritoneal lavage was performed using phosphate-buffered saline at pH 7.2. Cells were sedimented at 250 × g, and then cells were resuspended in DMEM-S at 1.5 × 106/ml. One

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hundred microliters of cell suspension was added to each well of a Costar (Cambridge, Mass.) 96-well tissue culture plate and allowed to adhere. Nonadherent cells were removed and cultures were treated as described below.

Cell culture. L929 cells were a gift from Sam Baron (Galveston, Tex.) and were cultured in DMEM-S in Corning T-75 tissue culture flasks. Cells were removed using 0.25% trypsin supplemented with EDTA (Mediatech, Herndon, Va.) and plated on Costar 96-well tissue culture plates at a concentration of 1.2 × 10⁵ to 1.5 × 10⁶ cells/ml. After adherence, cells were incubated with various doses of cocaine from 6 to 48 h.

Viral plaque assay. MHV strain MHV-JHM (ATCC VR-765) was obtained from the American Type Culture Collection (Rockville, Md.). The Indiana strain of vesicular stomatitis virus (VSV) was a gift from Sam Baron. The viral plaque assay has been described previously (9). Briefly, after Mβ and L929 cells were incubated with cocaine, the culture media were discarded and 50 µl of virus (MHV or VSV, respectively) diluted to approximately 40 PFU in DMEM-S was added. The cells were then incubated for 1 h at 37°C before 100 µl of overlay consisting of methylcellulose in DMEM-S was added. The plate was incubated until plaque formation occurred, and the monolayers were stained and fixed with 1% crystal violet (Sigma) in 80% methanol. The resulting viral plaques were counted, and a change in the number between control and treated wells was taken to indicate differences in overall virus replication.

Transfer of antiviral activity. To determine if there was a release and accumulation of antiviral products into the culture media of cells incubated with cocaine, L929 cells were grown to confluence on 96-well plates and incubated with 0 or 100 µg of cocaine per ml for 24 h. The culture media were then removed, the cells were washed to remove residual cocaine, and fresh media without cocaine were added for 6, 12, 24, or 48 h. At the times indicated below, the culture media were collected, samples were pooled, and 100 µl/well was transferred to fresh L929 cells for 24 h. After this incubation, the media were removed and a viral plaque assay with VSV was performed as described above.

To determine the identity of the antiviral products in the transferred supernatants from the above-described experiments, 2 ml of each sample was incubated with 100 µl of anti-IFN-α/β (2,500 U/ml) or nonimmune rabbit serum for 30 min before 100 µl/well was added to fresh L929 cells. A viral plaque assay was performed 24 h later.

Direct effects of antiserum. When the effects of IFN were determined directly on cocaine-exposed cells, the culture media were discarded after a 24-h incubation and 20 µl of anti-IFN (2,750 U of IFN-α/β per ml or 1,000 U of IFN-β per ml) in DMEM-S and cocaine were added. These cells not receiving antiserum were given 20 µl of control rabbit serum. After a 30-min incubation, a viral plaque assay was performed as described above except that the media in the wells were not discarded before virus was added.

IFN bioassay. L929 cells or Mβ were incubated with cocaine for 24 or 48 h, respectively. The culture media were then discarded and wells were washed to remove residual cocaine. One hundred microliters of a synthetic double-stranded RNA molecule, poly(I:C), at 1 or 5 µg/ml (Mβ and L929 cells, respectively) in DMEM-S and 100 µl of anti-IFN (2,750 U of IFN-α/β per ml) or nonimmune rabbit serum (9) were added for 1 h. L929 cells also received 200 µg of DEAE-dextran per ml to facilitate poly(I:C) incorporation. The cells were then washed twice to remove residual poly(I:C) and 100 µl of fresh media per ml was added for 6 h. The culture media were pooled and stored at −70°C until assayed. To assess the IFN activity in the samples, fresh plates of L929 cells were cultured to confluence and were later serial dilutions of samples were added for 18 h. A viral plaque assay with VSV was then performed as described earlier. The units of IFN were determined by taking the reciprocal of the dilution that inhibited the formation of 50% of the viral plaques.

RESULTS

Effect of cocaine on virus replication in L929 cells. When L929 cells were incubated with 0 to 100 µg of cocaine per ml, they exhibited increasing antiviral states as indicated by a dose-dependent reduction in plaque formation by VSV (Fig. 1). The maximum effect was observed at 100 µg/ml (approximately 300 µM), which was the highest concentration employed. A significant increase was noted when cells were incubated with as little as 3 µg/ml. To determine if cocaine’s effect was limited to VSV, L929 cells were incubated with 0 to 100 µg of cocaine per ml for 24 h and then a plaque assay was performed using MHV. The dose-dependent reduction of MHV plaque formation by cocaine was similar to the reduction seen with VSV (Fig. 1). L929 cells did not display a time-dependent difference in virus replication (data not shown) as noted previously with Mβ (9); however, a significant reduction in plaque number was noted after 6 h of incubation with cocaine. This was 18 h earlier than was observed for Mβ.

Transfer of antiviral activity. Direct exposure to cocaine has been shown to induce antiviral activity in both Mβ and L929 cells. Experiments were designed to determine if there was an accumulation of antiviral activity in the culture media of L929 cells incubated with cocaine. After incubation with 100 µg of cocaine per ml for 24 h, the cells were washed and fresh media without cocaine were added for 6, 12, 24, or 48 h. These cells displayed an antiviral effect for as long as 48 h after removal of cocaine, indicating that cocaine does not need to be present continuously (data not shown). When the culture media from the above-mentioned cells were transferred to fresh L929 cells, the samples displayed antiviral activity that was not evident until approximately 12 h after the removal of cocaine (Fig. 2). The maximal effect was observed at 24 h and began to diminish by 48 h.

When the transferred samples from the previous experiments were incubated with 2,500 U of rabbit anti-mouse IFN-α/β per ml before being placed on the fresh L929 cells, the antiviral activity was abolished, in contrast to that in samples incubated with nonimmune rabbit serum (Fig. 3).

Anti-IFN-α/β reverses cocaine’s effect. Because the previous experiments had shown that the addition of anti-IFN-α/β to transferred culture media from cocaine-treated cells blocked antiviral activity, experiments were performed to determine if
this effect could be produced on cells directly exposed to cocaine. L929 cells were incubated with 0 or 100 μg of cocaine per ml for 24 h. Culture media were then removed and replaced with media without cocaine for 6, 12, 24, or 48 h. Culture media were then collected and samples were transferred to fresh L929 cells for 24 h before a plaque assay with VSV was performed. n = 14 for all treatments. **, P < 0.01; ***, P < 0.005.

Effects of cocaine on poly(I:C)-induced IFN and TNF-α secretion in both L929 cells and Mφ. Because the antiviral activity of culture media from cocaine-treated cells was ablated by the presence of anti-IFN-α/β, experiments were conducted to quantify the increase in IFN secretion induced by cocaine. Both Mφ and L929 cells were incubated with different concentrations of cocaine for 24 to 48 h and then exposed to 1 or 3 μg of poly(I:C) per ml, respectively, for 6 to 8 h. The resulting culture medium samples were analyzed for IFN by bioassay. Cocaine caused an increase in the amount of IFN induced by poly(I:C) in both L929 cells and Mφ in a dose-dependent manner (Fig. 5). An enzyme-linked immunosorbent assay for TNF-α indicated that cocaine produced a dose-dependent decrease in TNF-α secretion from L929 cells and Mφ stimulated with poly(I:C) (data not shown).

Mφ incubated with 100 μg of cocaine per ml for 6, 12, 24, 48, or 72 h exhibited a time-dependent increase in the level of IFN secretion, with maximal levels being obtained when cells were incubated with cocaine for 48 h (Fig. 6). An increase in IFN secretion over that of control-treated cells was not observed until cells were incubated with cocaine for at least 24 h. An increase over time was not observed with L929 cells; however, a significant increase in IFN was noted after only 12 h. This was

FIG. 2. Secretion of an antiviral product by L929 cells incubated with cocaine. L929 cells were incubated with 100 μg of cocaine per ml for 24 h. Culture media were then removed and replaced with media without cocaine for 6, 12, 24, or 48 h. Culture media were then collected and samples were transferred to fresh L929 cells for 24 h before a plaque assay with VSV was performed. n = 14 for all treatments. **, P < 0.01; ***, P < 0.005.

FIG. 3. Anti-IFN-α/β ablates the antiviral activity in media from cocaine-exposed cells. L929 cells were incubated with 0 or 100 μg of cocaine per ml for 24 h before media were removed and 2,750 U of anti-IFN-α/β per ml in 20 μl of DMEM-S was added for 30 min. Control groups received 20 μl of control serum. A plaque assay with VSV was then performed without removing media already in the wells. n = 8 for all treatments. ***, P < 0.005.

FIG. 4. Anti-IFN-α/β reduces the antiviral activity in cells directly exposed to cocaine. L929 cells were incubated with 0 or 100 μg of cocaine per ml for 24 h. Culture media were then removed and 2,750 U of anti-IFN-α/β per ml in 20 μl of DMEM-S was added for 30 min. Control groups received 20 μl of control serum. A plaque assay with VSV was then performed without removing media already in the wells. n = 8 for all treatments. ***, P < 0.005.

FIG. 5. IFN responses of L929 and Mφ exposed to various levels of cocaine. L929 cells and Mφ were cultured with 0, 10, 33, or 100 μg of cocaine per ml for 24 and 48 h, respectively. Cells were then exposed to poly(I:C) at 1 μg/ml (Mφ) or 3 μg/ml (L929 cells). After 1 h, cells were washed and fresh media were added. Culture media were collected after 6 h, and IFN levels in the samples were determined by bioassay. n = 8 for all treatments. *, P < 0.05; ***, P < 0.005.
and cells were exposed to 1 nM walled and fresh media were added for 6 h. Samples were collected and ana-

\[ M_f \]

To determine if the increase in IFN secretion caused by cocaine was the result of an increase in mRNA level, an RNase protection assay was performed on L929 cells and \( M_f \). A dose-dependent increase in IFN transcript levels was observed, with average increases of two-fold over the control. The results of multiple experiments, and the average increase be-

\[ \text{stimulated with poly(I-C)} \]

and IFN-\( \alpha \) and IFN-\( \beta \) transcripts approximately four- and three-fold over control values, respectively. No IFN-specific transcripts were detected in \( M_f \) or L929 cells not stimulated with poly(I-C) (data not shown). Cocaine did not affect the production of constitutively expressed mRNA of GAPDH or L32.

FIG. 6. IFN response to \( M_f \) exposed to cocaine over time. \( M_f \) were incubated with 100 \( \mu \)g of cocaine per ml for 0, 12, 24, or 48 h. Media were removed and cells were exposed to 1 \( \mu \)g of poly(I-C) per ml for 1 h. Cultures were then washed and fresh media were added for 6 h. Samples were collected and ana-

\[ n = 8 \]

lyzed for IFN levels by bioassay. \( n = 8 \) for all treatments. **, \( P < 0.01 \); ***, \( P < 0.005 \).

consistent with the pattern of antiviral activity observed after direct exposure of cells to cocaine.

**Cocaine augments IFN transcript levels.** To determine if the increase in IFN secretion caused by cocaine was the result of an increase in mRNA level, an RNase protection assay was performed on L929 cells and \( M_f \) incubated with cocaine and stimulated with poly(I-C). Densitometry was performed for results of multiple experiments, and the average increase between control and cocaine-treated cells was determined. In \( M_f \), an average of a two-fold increase over the control was noted for IFN-\( \beta \) at the highest concentration of cocaine as indicated by densitometry (Fig. 7A). There was no expression of IFN-\( \alpha \) and only a small amount of IFN-\( \gamma \) transcript. In L929 cells, both IFN-\( \alpha \) and IFN-\( \beta \) were up-regulated (Fig. 7B). Densitometry indicated that 100 \( \mu \)g of cocaine per ml increased IFN-\( \alpha \) and IFN-\( \beta \) transcripts approximately four- and three-fold over control values, respectively. No IFN-specific transcripts were detected in \( M_f \) or L929 cells not stimulated with poly(I-C) (data not shown). Cocaine did not affect the production of constitutively expressed mRNA of GAPDH or L32.

**DISCUSSION**

The studies presented here offer a more thorough charac-
terization of cocaine’s antiviral effect. Cocaine produced a dose-dependent inhibition of plaque formation in L929 cells infected with either VSV or MHV. This supports our previous finding showing cocaine’s ability to inhibit MHV replication in \( M_f \). It also demonstrates that cocaine’s antiviral effect is not exclusive to any one cell type or virus and suggests that the effect is on a global antiviral product found in common in the different cells used in this study.

In previous experiments with \( M_f \), it was determined that antiviral activity could be transferred to fresh \( M_f \), indicating that cocaine-treated cells secreted antiviral products (9). How-

\[ \text{poly(I-C)} \]

ever, it was not determined what the identity of this secreted product was or whether it accumulated over time, which would be expected with the induction of secreted products. By incu-

\[ \text{poly(I-C)} \]

bating cells with cocaine for a static amount of time and then adding fresh media without cocaine for different amounts of time, we were able to show that the antiviral activity in the culture media did increase. This indicated that cocaine probably induced active release of an antiviral product into the media. It also suggests that cocaine is capable of stimulating antiviral secretion without the presence of another inducer [e.g., poly(I-C) or virus].

A possible explanation for the relatively small amount of antiviral activity detected in the transferred samples is that cocaine by itself is only a mild inducer of IFN secretion and cells do not constitutively produce IFN without a potent in-

\[ \text{poly(I-C)} \]

ducer. However, the stimulation by cocaine is multiplied by the presence of a potent IFN inducer like poly(I-C) or virus. Another possible explanation for the low antiviral activity in the transfer experiments is the necessity for removing cocaine during the latter stages of the experiment. Because the cells were cultured without cocaine stimulation for up to 48 h, the IFN induction would be expected to decrease. However, the re-

\[ \text{poly(I-C)} \]

moval of cocaine was necessary because L929 cells, which were being used to analyze antiviral products in transferred culture media, were also sensitive to cocaine’s effects.

The addition of anti-IFN-\( \alpha \)-\( \beta \) to either L929 cells or culture media collected from cocaine-exposed L929 cells reversed the antiviral activity of cocaine. These data are consistent with those obtained with \( M_f \) incubated with anti-IFN-\( \beta \). The finding that anti-IFN-\( \beta \) did not reverse the antiviral effect in L929 cells could be explained by the fact that these cells secrete as much as 40% of their IFN as IFN-\( \alpha \) (W. R. Fleischmann, Jr., University of Texas Medical Branch, Galveston, personal com-

\[ \text{poly(I-C)} \]

munication). It is also important to note that in these studies, murine \( M_f \) produced primarily IFN-\( \beta \), despite the fact that the literature defines them as being primarily a source of IFN-\( \alpha \). It was necessary to determine in a more quantitative manner what effects cocaine had on IFN secretion. When cells were incubated with different concentrations of cocaine and the IFN inducer poly(I-C), a dose-dependent increase in IFN was ob-

\[ \text{poly(I-C)} \]

served. A time-related increase was also seen in \( M_f \) but not L929 cells. These results are consistent with the antiviral re-

FIG. 7. Effects of cocaine on the level of poly(I-C)-induced RNA transcripts. \( M_f \) (A) or L929 cells (B) were incubated with cocaine at 0 or 100 \( \mu \)g/ml for 48 h. The cells were then cultured for 3 h with 1 or 3 \( \mu \)g of poly(I-C) per ml, respectively. Total cell RNA was collected and an RNase protection assay was performed as described in Materials and Methods. Densitometry was performed to determine differences between treatments and the loading of lanes. This assay was repeated at least twice to ensure reproducibility.
response of Mφ and L929 cells exposed to cocaine and then challenged with virus and IFN. Because TNF-α has been shown to have antiviral activity (23), experiments were performed to determine if part of cocaine’s inhibition of virus replication was due to an increase in TNF-α secretion, possibly by acting in synergy with IFN. However, the inhibition of TNF-α secretion by cocaine lends further support for IFN being the primary antiviral mechanism of cocaine.

The increase in IFN observed in these studies is supported by a report that acute cocaine exposure in vivo increased IFN-γ secretion for PBMC in cocaine-dependent human addicts (6). Also, Van Dyke et al. reported that human NK cell activity and distribution were increased after in vivo cocaine exposure (26). It has been reported that a major effect of IFN is to increase NK cell activity (2), thus leading to the possibility that the mechanism of cocaine’s effects on NK cells in that study was stimulation of IFN secretion.

An increase in secreted protein does not necessitate an increase in protein production. Instead, alterations in secretion or stability of the protein could affect the amount of product detected in the culture media. Therefore, an analysis of the amount of mRNA for several IFN genes was undertaken to determine if cocaine’s effects on IFN were at the transcriptional level. In an RNase protection assay, cocaine produced dose-related increases in mRNA for both IFN-α and IFN-β, with the greatest increase due to cocaine being observed in L929 cells. These results suggest that cocaine affects IFN secretion by increasing either the transcription of the IFN genes or the stability of the resulting transcripts. The fact that not all of the inducible transcripts detected in the assay were augmented supports transcriptional regulation. This is consistent with the literature, which states that the majority of IFN regulation occurs at the transcriptional level (25). The α species of IFN-α was chosen because it has previously been reported to be produced by L929 cells in larger amounts than the other IFN species (8). Neither of the two housekeeping genes was affected by cocaine, indicating that the increase in IFN mRNA levels was not due to an overall increase in transcription. The fact that no IFN-specific transcripts were detected in cells exposed to cocaine but not poly(I-C) could be a result of mRNA levels being below the detection limits of the assay. This is consistent with the fact that cells produce high levels of IFN only in the presence of an inducer like poly(I-C) or virus. This is further evidence that cocaine by itself induces only a slight stimulatory effect on IFN production. Also in support of this, previous research by other investigators, which has been replicated in this lab, has found that cocaine, in the absence of LPS or another stimulating agent, was insufficient to alter cytokine secretion (13).

IFN has been demonstrated to inhibit cell proliferation. Di Francesco et al. have reported that cocaine inhibited rat fibroblast proliferation; however, no mechanism was described (4). We have also observed that when L929 cells were treated with cocaine for at least 24 h, there was a slight but significant reduction in cellular proliferation that was reversible by the addition of anti-IFN antibodies (unpublished observation). It is possible that cocaine up-regulates the expression of baseline IFN message, thus resulting in a mild inhibition of cell proliferation.

The concentrations of cocaine found in murine serum after a subchronic schedule of cocaine administration range from 1 to 3 μg/ml within 30 min of the last dose (12). However, cocaine is rapidly metabolized in the body, resulting in various end products that have been found to be pharmacologically active (11, 17, 21). The tissue deposition of these metabolites varies considerably, making a precise measurement of exposure to cocaine and its metabolites difficult. However, 3 μg of cocaine per ml inhibited virus replication in the present study, suggesting that this effect could be of biological significance. A recent study by members of this group has reported a 50% decrease in influenza virus present in the lungs of mice receiving subchronic cocaine exposure (7). This is further support for a biologically relevant effect of cocaine on virus replication.

Other investigators have reported that cocaine induced an enhancement of viral replication. Peterson et al. reported that there was a dose-dependent increase in IFN replication in PBMC as indicated by an increased expression of the HIV p24 antigen (18). However, the overall replication of virus was never assessed. The increase could have been the result of enhanced protein secretion without a change in infectious virus production. It should be emphasized that the cocaine concentrations employed in the HIV studies were much lower (300 ng/ml) than those used in the present study. Also, the considerable differences between lymphocyte and Mφ responses could explain the discrepancies in results. Our data demonstrate that cocaine enhances IFN secretion in both murine peritoneal Mφ and L929 cells, but we have not yet assessed this phenomenon in a human cell line.

This study has demonstrated that cocaine has the capacity to increase the level of IFN secreted by murine thioglycolate-induced peritoneal Mφ and L929 cells and that at least part of the regulation of IFN by cocaine is on the level of transcript produced. These data correlate with the decrease in overall virus replication that has been reported in this and previous studies (9). The mechanism by which cocaine enhances IFN production remains unclear. However, we have noted an increase in intracellular calcium levels and mobilization in Mφ incubated with cocaine. As calcium has been shown to up-regulate gene transcription (16), it is possible that there exists a relationship between the augmentation of calcium and the increase in IFN production by cocaine; however, further research in this area is necessary.

REFERENCES


