Prostaglandins & other Lipid Mediators xxx (2015) xxx–xxx

Contents lists available at ScienceDirect

Prostaglandins and Other Lipid Mediators

Original Research Article

# N-Stearoylethanolamine suppresses the pro-inflammatory cytokines production by inhibition of NF-κB translocation

Andrey G. Berdyshev[a,*], Halyna V. Kosiakova[a,1], Oleksandra V. Onopchenko[a,1], Rostislav R. Panchuk[b,2], Rostislav S. Stoika[b,2], Nadiya M. Hula[a,1]

[a] Department of Lipid Biochemistry,[3] O.V. Palladin Institute of Biochemistry of National Academy of Science of Ukraine, Leontovich St. 9, 01601, Kyiv, Ukraine
[b] Department of Regulation of Cell Proliferation and Apoptosis, Institute of Cell Biology of National Academy of Science of Ukraine, [4] Drahomanov St. 14/16, 79005, Lviv, Ukraine

ARTICLE INFO

*Article history:*
Received 30 January 2015
Received in revised form 30 April 2015
Accepted 4 May 2015
Available online xxx

*Keywords:*
N-Stearoylethanolamine
NF-kB
Inflammation
Insulin resistance
Cytokines
N-Acylethanolamines

ABSTRACT

N-Stearoylethanolamine (NSE) is a minor lipid that belongs to the N-Acylethanolamines family that mediates a wide range of biological processes. This study investigates the mechanisms of anti-inflammatory action of NSE on different model systems. Namely, we estimated the effect of NSE on inflammatory cytokines mRNA level (leukemia cells L1210), cytokines content (serum and LPS-stimulated macrophages) and nuclear translocation of NF-κB (peritoneal macrophages LPS-stimulated and isolated from rats with obesity-induced insulin resistance). The results indicated that NSE dose-dependently inhibits the IL-1 and IL-6 mRNA level in L1210 cells. Furthermore, the NSE treatment triggered a normalization of serum TNF-α level in insulin resistant rats and a reduction of medium IL-1 level in LPS-activated peritoneal macrophages. These NSE's effects were associated with the inhibition of nuclear NF-κB translocation in rat peritoneal macrophages.



## 1. Introduction

N-Acylethanolamines (NAEs) are endogenous lipids that play an important role in numerous physiological and pathological processes by interacting with cannabinoid (CB) and noncannabinoid receptors. Anandamide is the most studied cannabinoid ligand that activates CB, TRPV (transient receptor potential channels) receptors and exerts analgesic, anti-inflammatory properties. In addition, anandamide shows orexigenic and lipogenic effect on energy metabolism [1]. Other N-acylethanolamides, with saturated (N-stearoylethanolamine (NSE), N-palmytoylethanolamine) or monounsaturated acyl chains (N-oleoylethanolamine) are thought to be CB-receptor inactive. The numerous studies suggest that saturated and monounsaturated NAEs are natural activators of peroxisome proliferator-activated receptors (PPARs). In particular, activation of the PPARα and γ isoforms has been shown to have anti-inflammatory and anorexic effects in a variety of tissues, which are likely to be connected with NAEs influence on NF-κB pathway [2].

Obesity is closely associated with a state of chronic, low-grade inflammation that causes the development of such world-spread diseases as insulin resistance, diabetes type 2 and cancer. Nowadays, so popular among youngsters fast-food causes a progression of abdominal obesity that reached the number of one and a half billion of people in the world, where more than 44% are people with diabetes [3]. High saturated fat overload induces hypertrophy of white adipose tissue that followed by macrophages activation and inflammatory cytokine production (TNFα, IL-6). Primarily overexpression of TNFα contributes to systemic dyslipidemia progression, insulin resistance and cancer growth "microenvironmental surrounding" development [4,5]. It was widely investigated that cytokines production induces phosphorylation (via activation of the IκB kinase complex) and subsequent proteasomal degradation of IκB inhibitory proteins, activating NFκB for nuclear translocation, where it promotes genes connected to pro-inflammatory mediators synthesis [6]. The PPARγ receptor subtype appears to play a pivotal

* Corresponding author. Tel.: +380 44 234 12 96.
*E-mail addresses:* kievlipids@gmail.com (A.G. Berdyshev), kosiakova@hotmail.com (H.V. Kosiakova), onop.89.av@mail.ru (O.V. Onopchenko), rpanchuk@ukr.net (R.R. Panchuk), stoika.rostyslav@gmail.com (R.S. Stoika), ngula@biochem.kiev.ua (N.M. Hula).
[1] Tel.: +380 44 234 12 96.
[2] Tel.: +380 32 2612108; fax: +380 32 2612148.
[3] E-mail address: secretar@biochem.kiev.ua.
[4] E-mail address: institut@cellbiol.lviv.ua.

http://dx.doi.org/10.1016/j.prostaglandins.2015.05.001
1098-8823/

role in the regulation of inflammation and dyslipidemia by blocking NF-κB nuclear translocation [7]. Previously, we have shown that NSE has anti-inflammatory action and accelerated the process of burn wound healing in the rat by the inhibition of cytokine IL-1β, IL-6 and TNFα production [8]. It worth to note that anti-inflammatory effect of N-palmitoylethanolamine (NPE) is widely studied, however, there is no experimental data about NSE.

Therefore, the aim of this study was to investigate the effect of NSE on inflammatory cytokine (IL-1β, IL-6 and TNFα) production in leukemia cell culture, serum and macrophages medium from rats with obesity-induced insulin resistance. In addition, it is important to determine whether this effect of NSE is associated with NF-κB nuclear translocation.

## 2. Materials and methods

### 2.1. Cell culture

The murine leukemic cells of the L1210 line were obtained from the Cell Culture Collection of the R. E. Kavetsky Institute of Experimental Pathology, Oncology and Radiobiology, National Academy of Sciences of Ukraine (Kyiv, Ukraine), maintained in a suspension culture consisting of Dulbecco's Minimum Essential Medium (Sigma Chemical Co., St. Louis, MO) supplemented with 10% heat-inactivated fetal calf serum (Sigma Chemical Co.) and gentamicin (50 μg/mL; Sigma Chemical Co.). The cells ($0.5 \times 10^6$ in each sample; $n = 3$) were seeded in 24-well plastic plates (Costar) and were incubated 24 h with NSE at different concentrations (10, 20 and 30 μM) at 37 °C in a humidified atmosphere of 5% $CO_2$ and 95% air.

### 2.2. Animal model

The study was carried out on male Sprague-Dawley rats ($188 \pm 38$ g). All procedures were conducted in accordance with the rules of the Commission on Bioethics of Institute of Biochemistry, National Academy of Sciences and with the "General ethical principles on experiments with animals" of the 1st National Congress on Bioethics (Kyiv, 2001). Rats were housed in standard cages with free access to food and water.

Obesity-induced IR was attained by feeding a prolonged high-fat diet (HFD) (58% fat: 23% proteins: 10% carbohydrates for 6 months) as described earlier [9]. The amount of lipids in the diet was increased by addition of lard to the pellet chow. The HFD fatty acid composition was at a ratio of 55% saturated (24% palmitic and 28% stearic acid) to 45% unsaturated fatty acid. Control rats during the experiment were on normal pellet diet (4% fat: 23% proteins: 65% carbohydrates) with saturated/unsaturated fatty acid ratio 38%/62%, respectively.

Six months after HFD period, we conducted the oral glucose tolerance test [10]. The results showed that after glucose administration (1 mL of 50% glucose solution) to HFD rats the level of blood glucose within 90 min was 8.5 mM (in controls – 5.1 mM), and within 150 min was higher than 5 mM (in controls – decreased to normal levels – 3.8 mM). The rats with impaired glucose tolerance were selected and divided into 2 groups: IR ($n = 9$) and IR + NSE ($n = 10$). Control rats with normal glucose tolerance were further subdivided into control ($n = 10$) and NSE ($n = 7$) groups. Animals in NSE and IR + NSE groups were orally received the water suspension of NSE for 2 weeks at the dose of 50 mg body weight $kg^{-1}$.

The establishment of IR was confirmed based on the results of fasting plasma insulin levels (measured by ELISA kit, DRG Germany) and HOMA-IR (homeostatic model assessment – insulin resistance) value, calculated by fasting insulin (nM) × fasting glucose (mM)/22.5. At the end of the experiment, the rats were scarified under Nembutal anesthesia (50 mg/kg body weight). The whole blood samples were collected and centrifuged. After that, serum was transferred into plastic tubes and frozen at −80 °C until further analysis.

### 2.3. Peritoneal macrophages isolation procedure

After scarification rat resident peritoneal macrophages were collected by lavaging the peritoneal cavity of rat ($n = 6$) with RPMI-1640 and cultivated directly on glass cover-slips in a 35 mm dish ($1.6 \times 10^6$ cells in each sample) with 10 μL lipopolysaccharide (LPS) solution (1 mg/mL PBS) and water suspension of NSE ($10^{-7}$ M).

### 2.4. Immunofluorescence confocal microscopy

The NF-κB activation was determined by immunofluorescence assays by evaluating the nuclear translocation of p-65 (NF-κB). Cells were fixed with 3.5% paraformaldehyde in PBS for 10 min at room temperature and permeabilized with 0.2% Triton X-100 (in PBS) for 5 min. To investigate the cellular localization of NF-κB, cells were treated with a polyclonal antibody (1:600) against NF-κB p65 (Sigma–Aldrich) for 1.5 h. After extensive washing with PBS, cells were further incubated with a secondary FITC-conjugated donkey anti-rabbit IgG antibody (Sigma–Aldrich) diluted at 1:500 in PBS for 1 h at a room temperature. Nuclei were stained with 0.5 μg/mL of DAPI (Sigma–Aldrich), and then analyzed by confocal microscopy using a Zeiss LSM 510 Meta microscope. Randomly, 30–150 cells of each confocal image slide ($1024 \times 1024$ pixels) were counted with independently developed software based on ImageEn component suite (Xequte Software, New Zealand) that calculated the percentage of green pixels in the nucleus zone. The percentage of FITC particles (p65 subunit of NF-κB) was equal to green pixels.

### 2.5. Cytokine level measurement

The IL-1 level in culture supernatants and TNF-α, IL-6, IL-1 level in rat serum were measured by the respective ELISA kits (eBioscience, Austria).

### 2.6. RT-PCR analysis

RT-PCR analysis was used for evaluating the expression of mRNA coding for several cytokines. Total RNA was isolated from L1210 cells using Trizol reagent (Sigma, USA). mRNA ($A_{260}/A_{280} = 1.8 \pm 0.1$) was converted to cDNA using "RevertAidTM First Strand cDNA Synthesis Kit" (Fermentas, Lithuania). The obtained cDNAs were subjected to RT-PCR analysis using specific primers for murine cytokine cDNAs (Table 1).

### 2.7. Gene primer sequence

Optimal conditions for RT-PCR analysis were chosen experimentally: the temperature was 94, 55 and 72 °C ($n = 30$) for cytokine cDNA, and 94, 65 and 72 °C ($n = 25$) for β-actin cDNA, used as a reference gene. It is known, that β-actin belongs to a family of

**Table 1**
Primers used in the study.

| | |
|---|---|
| IL-1α | |
| | Forward 5′-GCCAGTTGAGTAGGATAAAGG-3′ |
| | Reverse 5′-CAGTCTGTCTCCTTCTTGAGG-3′ |
| IL-6 | |
| | Forward 5′-TGGAGTCACAGAAGGAGTGGCTAAG-3′ |
| | Reverse 5′-TCTGACCACAGTGAGGAATGTCCAC-3′ |
| β-Actin | |
| | Forward 5′-TCACCCACACTGTGCCCATCTA-3′ |
| | Reverse 5′-CAGCGGAACCGCTCATTGCCAA-3′ |

Please cite this article in press as: Berdyshev AG, et al. N-Stearoylethanolamine suppresses the pro-inflammatory cytokines production by inhibition of NF-κB translocation. Prostaglandins Other Lipid Mediat (2015), http://dx.doi.org/10.1016/j.prostaglandins.2015.05.001

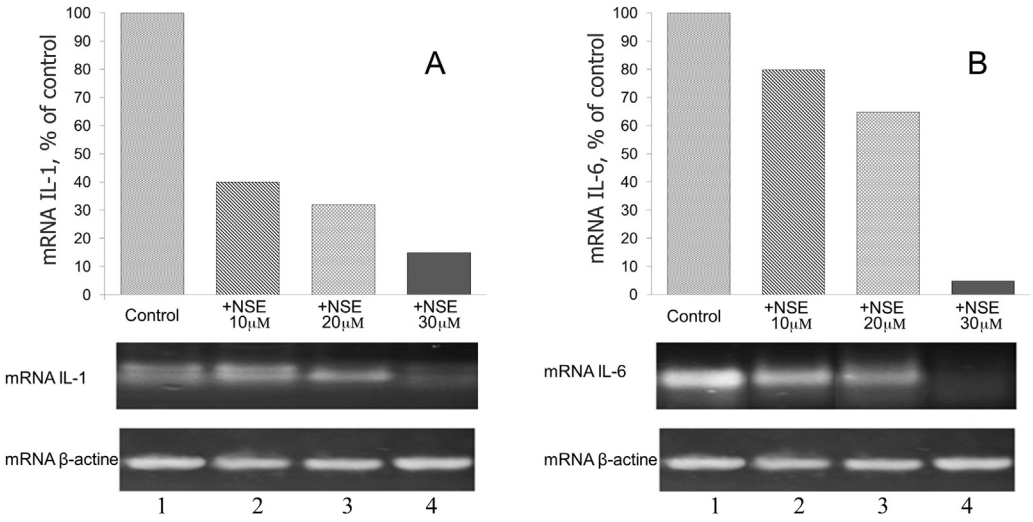


**Fig. 1.** Effect of NSE on IL-1 (A) and IL-6 (B) production in L1210 cells. The level of cytokines mRNA were determined by RT-PCR analysis. IL, interleukin; NSE, N-stearoylethanolamine.

housekeeping genes whose expression is assumed to be stable and does not differ in various types of cells.

The DNA products of PCR reaction were resolved in 1% agarose gel, visualized by ethidium bromide, and photographed by an Olympus C-5000 digital camera. The DNA bands were evaluated quantitatively by using GelPro Analyzer 3.1 software.

### 2.8. Data processing method

Data processing was performed by calculating the average and standard deviation in the measurements for each group using the Windows statistical program SPSS 18.0. Repeated measures two-way (ANOVA) analysis was used to analyze the differences in the groups, time and to identify group × time interactions. The *t*-test for matching samples was performed to verify the differences before and after exercise within the exercise group for the interaction analysis. The significant level of all the statistics data is $\alpha = 0.05$.

## 3. Results

### 3.1. Cytokines production in L1210 cells

The incubation of L1210 cells with NSE showed dose-dependent inhibition of IL-1 (Fig. 1A) and IL-6 (Fig. 1B) production. The most effective NSE dose was 30 μM, where IL-1, IL-6 mRNA expression were 70% and 95% lower in comparison with control cells.

### 3.2. Animal weight, plasma insulin level and index HOMA-IR value

The initial weight of rats from "Control" and "IR" groups was 188 ± 38 g. Throughout the experiment, the rats were gaining weight gradually and at the 24th week the final weigh of HFD rats was 420 ± 10 g (due to visceral fat) in comparison with control rats of 340 ± 15 g. After the HFD overload, the rat plasma insulin level was almost 3-fold higher than in "Control" group (Table 2). The estimation of HOMA-IR in "IR" group of rats showed significant increase in its value compare to controls (Table 2). Meanwhile, in IR rats that were treated by NSE the plasma level of insulin was 20% lower than those of "IR" group and was accompanied by a reduction of HOMA-IR value (Table 2).

### 3.3. Rat serum cytokine content

Our results demonstrate that TNF-α level was 3-fold higher in serum of rats from IR group compared to Control (Fig. 2A). The administration of NSE normalized the TNF-α serum content in IR rats. We found no significant changes in control rats that were treated by NSE.

Meanwhile, we have not found any considerable differences in serum IL-1 and IL-6 level (Fig. 2B and C).

### 3.4. Cytokine level in culture medium of rat LPS-activated peritoneal macrophages

In our study the LPS activation of peritoneal macrophages caused the increase of IL-1 (Fig. 3) level. The incubation of macrophages with NSE resulted in normalization of this cytokine content.

### 3.5. NF-κB nuclear translocation in the macrophages

We found that in controls the percentage of luminescence intensity in the nucleus was 20–40 (Fig. 4A). In control macrophages stimulated by LPS (Fig. 4A) and macrophages isolated from IR rats (Fig. 4B) the percentage of FITC particles was increased in the nucleus to 60–70 and 70–80, respectively. The macrophages isolated from IR rats showed the same level of nuclear FITC particles as in IR macrophages after LPS stimulation that confirms activation of peritoneal macrophages under obesity-induced IR. The incubation of LPS-stimulated control macrophages and macrophages from IR rats with NSE shows the reduction of FITC particles percentage in the nucleus to 20–30 (Fig. 4A) and to 50 (Fig. 4B), respectively. In Fig. 4C a summarizing bar chart of FITC particles percentage is presented.

**Table 2**
Plasma insulin level and HOMA-IR value in rat. Values are presented as mean ± SEM ($n = 6$–10/group). $^{*}P < 0.05$, compared to the "Control" group; $^{\#}P < 0.05$, compared to the "IR" group. IR, insulin resistance; NSE, N-stearoylethanolamine.

| | Control | IR | IR + NSE |
|---|---|---|---|
| Insulin (nM) | 2.3 + 0.09 | 6.36 + 0.4* | 5.1 + 0.4*,# |
| HOMA-IR (e.u.) | 0.38 + 0.01 | 1.34 + 0.12* | 0.75 + 0.04# |

Please cite this article in press as: Berdyshev AG, et al. N-Stearoylethanolamine suppresses the pro-inflammatory cytokines production by inhibition of NF-κB translocation. Prostaglandins Other Lipid Mediat (2015), http://dx.doi.org/10.1016/j.prostaglandins.2015.05.001

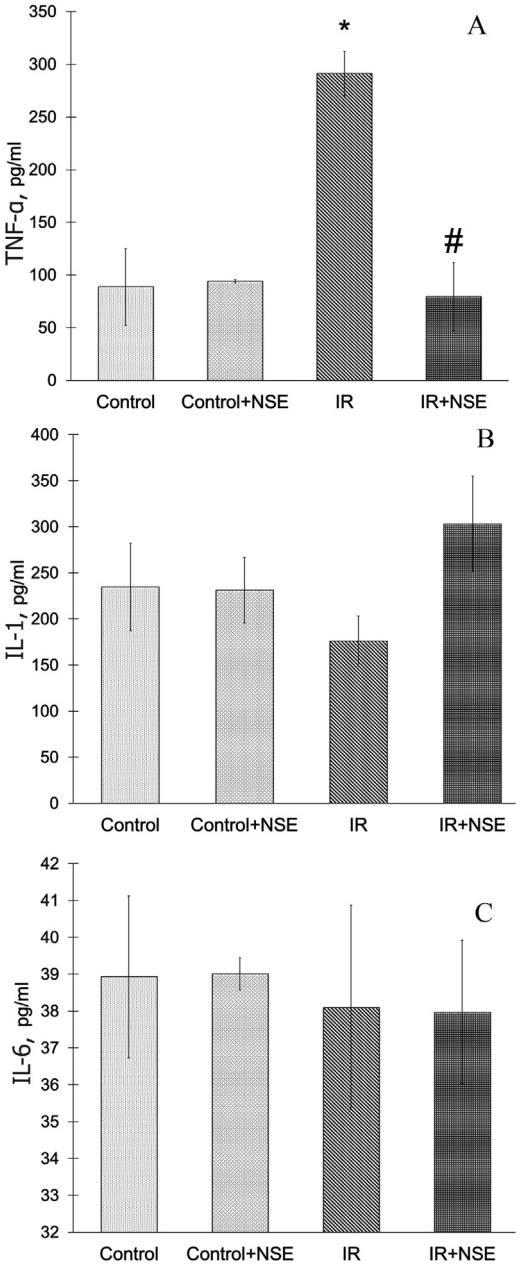


**Fig. 2.** Serum content of TNF-α (A), IL-1 (B) and IL-6 (C) in rat. Values are presented as mean ± SEM ($n$ = 6–10/group). *$P$ < 0.05, compared to the "Control" group; #$P$ < 0.05, compared to the "IR" group. IL, interleukin; IR, insulin resistance; NSE, N-stearoylethanolamine; TNF-α, tumor necrosis factor-α.

## 4. Discussion

The inflammation is thought to be the main and common factor in the development of such pathologies as diabetes, cancer and cardiovascular diseases. It is known that primarily cytokines (TNF-α, IL-1 (α,β), IL-6) initiate inflammatory process and are involved in its progression. Previously, the investigation of biological activity of NSE has shown its anti-inflammatory action at burn trauma in rats that was followed by faster wound healing, reduction of inflammatory cytokines level and restoration of pro-/antioxidant balance [8].

In the following study we investigated whether NSE would show its anti-inflammatory action at other different model systems. Thus, we studied the effect of NSE on the production of IL-6, IL-1 in murine leukemic cells and found that these cytokines production

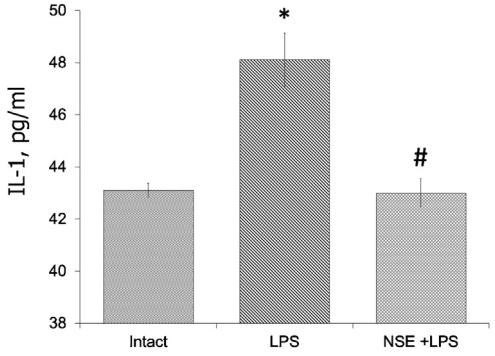


**Fig. 3.** Level of IL-1 in the culture supernatants of intact and LPS-activated rat peritoneal macrophages. Values are presented as mean ± SEM ($n$ = 6/group). *$P$ < 0.05, compared to intact macrophages; #$P$ < 0.05, compared to LPS-activated macrophages. LPS, lipopolysaccharide; NSE, N-stearoylethanolamine.

was considerably decreased in cells that were treated by different concentrations of NSE. Recent data showed the crosslink between inflammation and leukemia progression. Namely, human IL-1 induced the production of stem cell factor, which is a major hematopoietic growth factor that controls the progression of acute myeloid leukemia upon malignant transformation of hematopoietic myeloid cells [11]. Therefore, we propose that the decrease of IL-1 production under the NSE treatment may influence the growth and proliferation of leukemic cells.

The LPS stimulation of macrophages induces the elevated production of IL-1, TNF-α, IL-6, which trigger the inflammatory response [12]. Our findings represented that the pre-incubation of rat peritoneal macrophages with NSE resulted in a significant decrease of IL-1 level after theirs LPS activation. According to this, we suggest that NSE is able to suppress the IL-1 production that is in agreement with the results obtained in L1210 cells.

It is well known that structural analogue of NSE–N-palmitoylethanolamine (NPE) exerts anti-inflammatory activity by inhibition of inflammatory cytokines production via PPARα activation [13,14]. However, the earlier data presented no effect of NPE on the IL-6 secretion and NF-κB translocation in the human adipocytes [15]. Moreover, later results showed the inhibition by NPE of the LPS-induced secretion of TNF-α in the human adipocytes that was not associated with PPARα activation [16]. Unfortunately, now there is no information about the effect of NPE on cytokines synthesis in activated macrophages.

From investigations, under different pathological conditions the ratio of endogenous NAEs significantly changes, particularly, analysis of human muscle tissue showed almost two-fold higher level of NSE compared to NPE in myalgic subjects [17]. This finding suggests an important role of NSE in regulation of inflammatory response. In our experiment, at a rat model of HFD-induced insulin resistance we found that NSE reduces serum TNF-α level. Considering that TNF-α is primarily produced by NF-κB-dependent mechanism in activated macrophages, we studied the effect of NSE on the NF-κB translocation in the nucleus of isolated peritoneal macrophages from IR rats. It is worth to note, that the macrophages from rats with IR were already activated and the further incubation with LPS triggered no significant change. The obtained results showed that the treatment by NSE of macrophages isolated from IR rats triggered an inhibition of NF-κB translocation in the nucleus. Nowadays it is known that NSE does not activate PPARα, however, we suggest that its biological properties may be realized via PPARγ. Recent study indicate that up-regulation of heme oxygenase-1 by PPARγ signaling activation promotes differentiation of macrophages to anti-inflammatory phenotype and caused an alleviation of the inflammatory pain development [18]. In addition, the mice knockout on macrophage PPARγ have demonstrated

Please cite this article in press as: Berdyshev AG, et al. N-Stearoylethanolamine suppresses the pro-inflammatory cytokines production by inhibition of NF-κB translocation. Prostaglandins Other Lipid Mediat (2015), http://dx.doi.org/10.1016/j.prostaglandins.2015.05.001

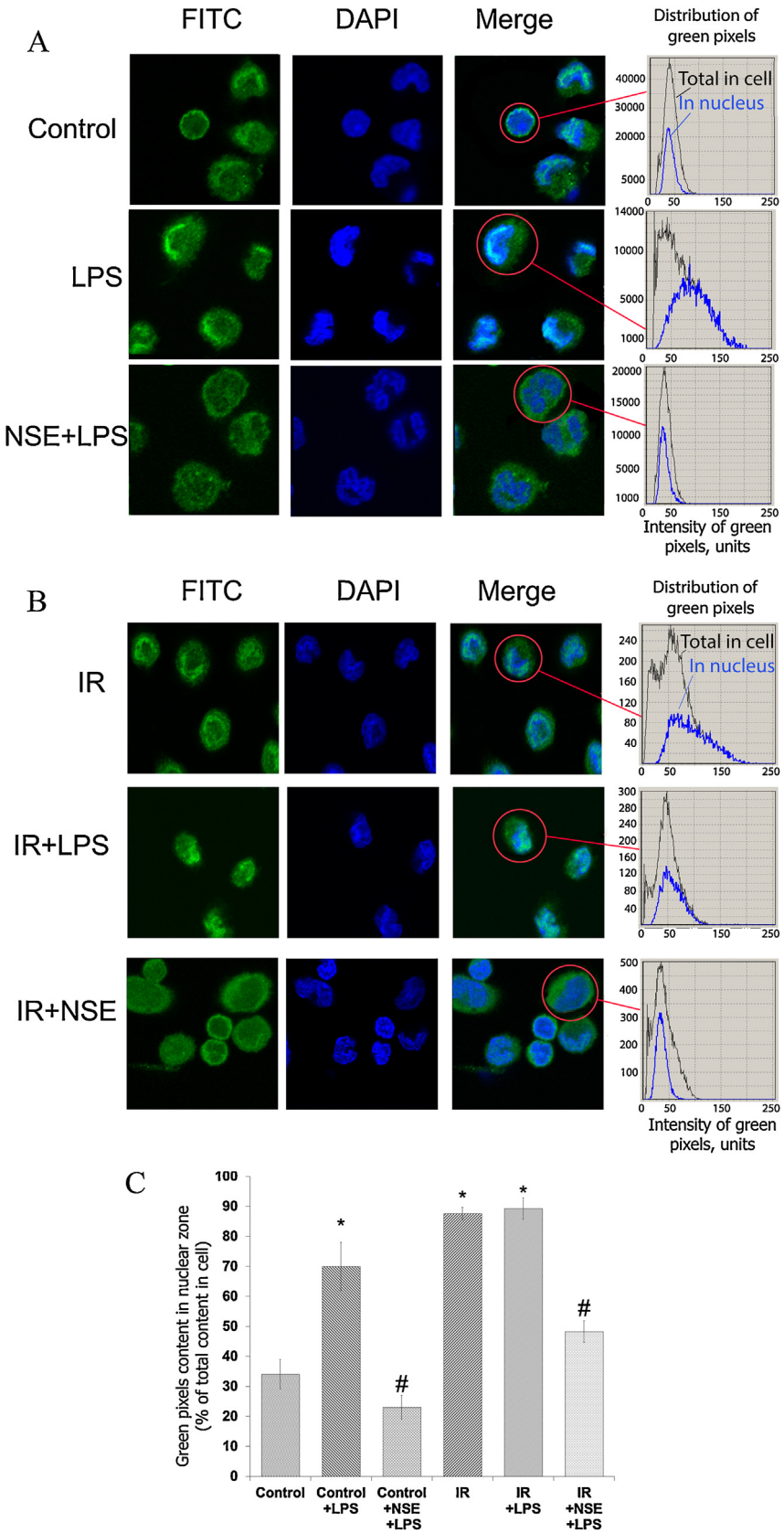


**Fig. 4.** Distribution of FITC-labeled IgG antibody (P65) NF-κB in peritoneal macrophages from control (A) and IR (B) rats. (C) Graphical representation of % FITC-labeled IgG antibody (P65) NF-κB in the nucleus of peritoneal macrophages (summarizing bar chart). Values are presented as mean ± SEM. Values in "Control" ($n = 72$), "Control + LPS" ($n = 87$), "Control + NSE + LPS" ($n = 65$), "IR" ($n = 48$), "IR + LPS" ($n = 53$), "IR + NSE + LPS" ($n = 79$) were comparable. $^{*}P < 0.05$, compared to the "Control" group; $^{\#}P < 0.05$, compared to the "Control + LPS" group. IR, insulin resistance; LPS, lipopolysaccharide; NSE, N-stearoylethanolamine.

Please cite this article in press as: Berdyshev AG, et al. N-Stearoylethanolamine suppresses the pro-inflammatory cytokines production by inhibition of NF-κB translocation. Prostaglandins Other Lipid Mediat (2015), http://dx.doi.org/10.1016/j.prostaglandins.2015.05.001

heightened inflammatory pathway activation, reduced glucose intolerance and insulin resistance on normal and high fat diets [19]. In our previous experiments at a model of obesity-induced insulin resistance we have shown the corrective effect of NSE on pancreas, liver dyslipidemia and improvement of insulin sensitivity [20,21]. It is known that PPARγ regulates lipid metabolism in different tissues and its activation by exogenous agonists causes a normalization of lipid composition and restoration of sensitivity to insulin [22].

Taking these findings into consideration, we suggest that biological activity of NSE may be realized via PPARγ modulation. The preponderance of evidence points to a crosstalk between PPARγ and transcription factor NF-κB (trans-repression) [23,24]. Modulation of this pathway by NSE may be used in inhibition of the inflammation/insulin resistance axis in diabetes type 2 complications. Therefore, further investigations into the role of NSE on PPARγ activation are necessary for better understanding of signaling mechanisms underlying the anti-inflammatory and insulin-sensitizing effect of NSE.

## 5. Conclusions

The represented results show that the less studied member of NAE's family, NSE exerts anti-inflammatory action by down-regulation of the pro-inflammatory cytokines expression and reduction of theirs level. This NSE's effect is associated with the inhibition of NF-κB translocation into the nucleus that was shown in isolated from insulin resistant rats' peritoneal macrophages. Therefore, we suggest that NSE is a perspective anti-inflammatory agent and may be used in pharmacotherapy of inflammatory-associated diseases.

## Conflict of interest

The authors declare no conflict of interest.

## Acknowledgments

This study was supported by the grants from National Academy of Sciences of Ukraine (№ 0110U005964) and (2.2.10 №6).

## References

[1] Matias I, Gonthier M-P, Petrosino S, Docimo L, Capasso R, Hoareau L, Monteleone P, Roche R, Izzo AA, Di Marzo V. Role and regulation of acylethanolamides in energy balance: focus on adipocytes and b-cells. Br J Pharmacol 2007;152:676–90.
[2] Sun Y, Alexander SPH, Kendall DA, Bennet AJ. Cannabinoids and PPARα signalling. Biochem Soc Trans 2006;34:1095–7.
[3] New data highlight increases in hypertension, diabetes incidence. Fact sheet No. 311; 2014. http://www.who.int/mediacentre/factsheets/fs311/en/ World health organization [accessed January 2015].
[4] Wellen KE, Hotamisligil GS. Obesity-induced inflammatory changes in adipose tissue. J Clin Invest 2003;112:1785–8.
[5] Osório-Costa F, Carvalheira JBC. TNF-α in obesity-associated colon cancer. Transl Gastrointest Cancer 2013;2:179–93.
[6] Zhou Y, Eppenberger-Castori S, Eppenberger U, Benz CC. The NFkB pathway and endocrine-resistant breast cancer. Endocrine-Related Cancer 2005;12:37–46.
[7] Ruan H, Pownall HJ, Lodish HF. Troglitazone antagonizes tumor necrosis factor-alpha-induced reprogramming of adipocyte gene expression by inhibiting the transcriptional regulatory functions of NF-kappaB. J Biol Chem 2003;278:28181–92.
[8] Zhukov AD, Berdyshev AG, Kosiakova GV, Klimashevskiy VM, Gorid'ko TM, Meged OF, Hula NM. N-stearoylethanolamine effect on the level of 11-hydroxycorticosteroids, cytokines IL-1β, IL-6 and TNFα in rats with nonspecific inflammation caused by thermal burn of skin. Ukr Biochem J 2014;86: 88–97.
[9] Svegliati-Baroni G, Candelaresi C, Saccomanno S, Ferretti G, Bachetti T, Marzioni M, De Minicis S, Nobili L, Salzano R, Omenetti A, Pacetti D, Sigmund S, Benedetti A, Casini A. A model of insulin resistance and nonalcoholic steatohepatitis in rats: role of peroxisome proliferator-activated receptor-alpha and n-3 polyunsaturated fatty acid treatment on liver injury. Am J Pathol 2006;169: 846–60.
[10] Onopchenko OV, Kosiakova HV, Horid'ko TM, Berdyshev AH, Mehed' OF, Hula NM. The effect of N-stearoylethanolamine on the activity of antioxidant enzymes, content of lipid peroxidation products and nitric oxide in the blood plasma and liver of rats with induced insulin-resistance. Ukr Biokhim Zh 2013;85:88–96.
[11] Wyszynski RW, Gibbs BF, Varani L, Iannotta D, Sumbayev VV. Interleukin-1 beta induces the expression and production of stem cell factor by epithelial cells: crucial involvement of the PI-3 K/mTOR pathway and HIF-1 transcription complex. Cell Mol Immunol 2014; http://dx.doi.org/10.1038/cmi.2014.113
[12] Tracey KJ. The inflammatory reflex. Nature 2002;420:853–9.
[13] Berdyshev E, Boichot E, Corbel M, Germain N, Lagente V. Effects of cannabinoid receptor ligands on LPS-induced pulmonary inflammation in mice. Life Sci 1998;63:125–9.
[14] Di Paola R, Impellizzeri D, Torre A, Mazzon E, Cappellani A, Faggio C, Esposito E, Trischitta F, Cuzzocrea S. Effects of palmitoylethanolamide on intestinal injury and inflammation caused by ischemia-reperfusion in mice. J Leukoc Biol 2012;91:911–20.
[15] Hoareau L, Ravanan P, Gonthier MP, Delarue P, Gonçalves J, Césari M, Festy F, Roche R. Effect of PEA on LPS inflammatory action in human adipocytes. Cytokine 2006;34:291–6.
[16] Hoareau L, Buyse M, Festy F, Ravanan P, Gonthier MP, Matias I, Petrosino S, Tallet F, d'Hellencourt CL, Cesari M, Di Marzo V, Roche R. Anti-inflammatory effect of palmitoylethanolamide on human adipocytes. Obesity (Silver Spring) 2009;17:431–8.
[17] Ghafouri N, Ghafouri B, Larsson B, Turkina MV, Karlsson L, Fowler CJ, Gerdle B. High levels of N-palmitoylethanolamide and N-stearoylethanolamide in microdialysate samples from myalgic trapezius muscle in women. PLoS ONE 2011;6:e27257.
[18] Hasegawa-Moriyama M, Kurimoto T, Nakama M, Godai K, Kojima M, Kuwaki T, Kanmura Y. Peroxisome proliferator-activated receptor-gamma agonist rosiglitazone attenuates inflammatory pain through the induction of heme oxygenase-1 in macrophages. Pain 2013;154:1402–12.
[19] Hevener AL, Olefsky JM, Reichart D, Nguyen MT, Bandyopadyhay G, Leung HY, Watt MJ, Benner C, Febbraio MA, Nguyen AK, Folian B, Subramaniam S, Gonzalez FJ, Glass CK, Ricote M. Macrophage PPARγ is required for normal skeletal muscle and hepatic insulin sensitivity and full antidiabetic effects of thiazolidinediones. J Clin Investig 2007;117:1658–69.
[20] Onopchenko OV, Kosiakova GV, Oz M, Klimashevsky VM, Gula NM. N-Stearoylethanolamine restores pancreas lipid composition in obesity-induced insulin resistant rats. Lipids 2015;50:13–21.
[21] Onopchenko OV, Kosiakova HV, Horid'ko TM, Klimashevskyi VM, Hula NM. The effect of N-stearoylethanolamine on liver phospholipid composition of rats with insulin resistance caused by alimentary obesity. Ukr Biokhim Zh 2014;86:101–10.
[22] Monsalve FA, Pyarasani RD, Delgado-Lopez F, Moore-Carrasco R. Peroxisome proliferator-activated receptor targets for the treatment of metabolic diseases. Mediators Inflamm 2013;2013:549627.
[23] Ricote M, Glass CK. PPARs and molecular mechanisms of transrepression. Biochim Biophys Acta 2007;1771:926–35.
[24] Olefsky JM, Glass CK. Macrophages, inflammation, and insulin resistance. Annu Rev Physiol 2010;72:219–46.