Analytical and anti-inflammatory effectiveness of sitagliptin and vildagliptin in mice

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ABSTRACT

To validate the potential anti-inflammatory and analgesic role of sita- and vildagliptin, five different experimental models were used in mice: i) mustard oil-induced ear edema, ii) neutrophil accumulation, iii) mechanical and iv) thermal touch sensitivity in complete Freund’s adjuvant-induced arthritis and v) capsaicin-induced plasma extravasation in the urinary bladder. For the complete examination period in i) the dose of 10 mg sitagliptin as well as 1–10 mg vildagliptin was found to significantly decrease ear edema as compared to positive control (p < 0.05, n = 8/group). All doses of sitagliptin provided an anti-inflammatory effect (p < 0.005 (n = 10/group) in test ii) and an analgesic effect in iii) except 3 mg. Vildagliptin was similarly effective in test ii) (p < 0.005, n = 10/group) as sitagliptin, but it failed to affect mechanical touch sensitivity. Unlike mechanical touch sensitivity, both gliptins could beneficially act on the thermal threshold (p < 0.05, n = 10/group). And 30 only in tests v) could both gliptins reverse inflammation. Further studies are needed to support the suggestion that the utilization of these beneficial effects of gliptins may be considered in the treatment of Type 2 diabetic patients.

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1. Introduction

Chronic inflammation and pain can be highly debilitating. To reduce the inflammation itself or to relieve the related pain is a justifiable expectation of the patients. Anti-inflammatory and analgesic drugs are commonly prescribed for the symptomatic treatment of different diseases and the range of chemical classes of available drugs is quite broad. The most frequently used drugs are the non-steroidal anti-inflammatory drugs, although the application of steroid compounds in serious cases is also widely accepted. The conditions when these drugs are applied are mostly immune-driven diseases like multiple sclerosis, inflammatory bowel disease, or rheumatoid arthritis. Moreover, diabetes related pain such as diabetic neuropathy or painful diabetic neuritis afflict a majority of diabetic patients especially, if the diabetes is not treated adequately.

Since diabetes (especially type-2 diabetes) has a growing prevalence worldwide, novel treatments of the disease are in the focus of scientific interest. The two most recently accepted incretin mechanisms involving drug categories are the degradation-resistant glucagon-like peptide-1 (GLP-1) receptor agonists (incretin mimetics) and the inhibitors of dipeptidyl peptidase-4 (DPP-4) activity (incretin enhancers) [1]. The pharmacological actions of GLP-1 analogues and DPP-4 inhibitors have been reviewed recently [2].

There are intestinal hormones released after the oral administration of glucose. These hormones are released in a glucose-dependent manner and are responsible for augmenting insulin secretion, promoting β cell proliferation and reducing apoptosis. This is defined as the incretin effect. The two most important hormones involved in the incretin mechanism are the glucose-dependent insulinotropic polypeptide (GIP) and GLP-1 [1,3]. Both GIP and GLP-1 are rapidly inactivated after their release; the half-life of active GLP-1 being less than 2 minutes. The inactivation is caused by a truncation of the peptides by the removal of the N-terminal peptide end. This process is executed by the enzyme dipeptidyl peptidase-4 (DPP-4) [4]. DPP-4 is a 110-kDa type-II integral membrane glycoprotein with ubiquitous expression and whose enzyme activity has been recorded in rats, mice and humans. It is present in the epithelial cells of the intestine, kidney, liver, lung, thymus, lymph node, spleen, prostate and in adipocytes, as well as on activated lymphocytes and monocytes [5]. Besides the incretin hormones, a number of bioactive peptides may be considered in the treatment of Type 2 diabetic patients.

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adenylate-cyclase-activating polypeptide, insulin-like growth factor-1, substance P and various chemokines [6]. DPP-4 is also known as the cell surface antigen CD26 and it can associate with the lymphocyte cell-surface molecules CD45 and adenosine deaminase (ADA) to have a co-stimulatory function in the immune response [5]. An interesting observation is the increase in the plasma concentration of DPP-4 as a soluble protein during continuous treatment of humans by sitagliptin (100 mg/day). This might originate from shedding of CD26 proteins from mononuclear cells evoked by sitagliptin [8].

Dipeptidyl peptidase-4 inhibitors, like sitagliptin and vildagliptin, have been already introduced to the market since 2006 and are used for the treatment of type-2 diabetes. Gliptins are found to improve the vascular endothelial function, thus performing pleiotropic cardiovascular actions [7]. The safety of the gliptin family was questioned recently, but in two long-term cardiovascular outcome trials, Saxagliptin Assessment of Vascular Outcomes Recorded in Patients with Diabetes Mellitus–Thrombolysis in Myocardial Infarction 53 (SAVOR-TIMI 53), it has been proven that saxagliptin is safe from the cardiovascular point of view. It was shown that the primary endpoints of the study (a composite of cardiovascular death, non-fatal myocardial infarction or non-fatal ischemic stroke) occurred in 7.3% of the saxagliptin group compared with 7.2% of the placebo group (ClinicalTrials.gov Identifier: NCT01107886). The conclusion of Cardiovascular Outcomes Study of Alogliptin in Patients With Type 2 Diabetes and Acute Coronary Syndrome (EXAMINE) study (ClinicalTrials.gov Identifier: NCT00968708) was that in type-2 diabetic patients with recent acute coronary syndrome, major cardiovascular event rates for alogliptin were not increased compared to placebo. In this trial acute pancreatitis development as a serious adverse event was only 0.07% compared to placebo (0.15%), thus it is valid to state that alogliptin is free from this side effect.

Both incretins, GIP and GLP-1 stimulate insulin secretion in a glucose dependent manner and consequently, DPP-4 inhibitor treatment does not increase the risk of hypoglycaemia. Not only was the occurrence of hypoglycaemic events incidentally similar or lower when comparing groups treated with DPP-4 inhibitor (either monotherapy or in combination) with placebo treated groups in different studies, but the number of reported adverse events did not differ from the actively treated groups. [4]. It has been demonstrated in animal studies that toxicity may be caused by the inhibition of other enzymes in this family, like DPP-8 and DPP-9 [9], so the selectivity of inhibitors to DPP-4 is crucially important to ensure an optimal safety profile. Since both sitagliptin and vildagliptin show a higher relative selectivity for DPP-4, the risk of development of adverse effects due to inhibition of other enzymes is minimized [4,10]. However, it did turn out that during the post marketing period of gliptins these DPP-4 inhibitors increased the rate of infections such as nasopharyngitis and urinary tract infections [11]. In addition, pancreatitis was reported mainly associated with the use of sitagliptin and linagliptin [12], although a recent meta-analysis could not find differences between DPP-4 inhibitors [13]. In spite of the increased risk of infections, sitagliptin and vildagliptin are well tolerated in general. Besides the primary targeted therapeutic area, in vitro and in vivo studies showed anti-inflammatory properties of DPP-4 inhibitors that could lead to a novel drug class for anti-inflammatory disorders [14]. Altered circulating peptidase activity and membrane DPP-4 expression have been demonstrated in a number of human inflammatory diseases [15]. DPP-4 is responsible for the metabolism of a number of regulatory factors, such as peptides or chemokines and affects the signaling functions. This suggests that DPP-4 is involved in determining immune response and procession of inflammatory disorders as well. As mentioned previously, DPP-4 is also known as the cell surface antigen CD26, which signals T-cells to proliferate. However, this mechanism cannot be attributed to the DPP-4 inhibition [16] because the T-cell activation seems to be independent of the DPP-4 enzyme activity and the ADA-binding capability [16,17]. Moreover, reversible DPP-4 inhibitor Lys[Z(NO2)]-pyrrolidide was shown to suppress autoimmune encephalomyelitis and upregulated TGF-β1 secretion in vivo [18].

The possible anti-inflammatory property of the gliptin group can be considered as an additional value of these drugs in diabetic patients with neuritis or diabetic neuropathy, or patients with atherosclerosis considering that these diseases are driven by inflammatory processes [19]. Moreover, the reduction in plasma C-reactive protein concentration and systolic blood pressure have been described for exenatide [20]. The anti-inflammatory action of sitagliptin [8] and exenatide [19] are proven biochemically in humans, thus in the present series of experiments we aimed to examine the possible anti-inflammatory effect of two potent DPP-4 inhibitors, sitagliptin and vildagliptin. They were applied in in vivo inflammation and analgesic models in mice.

2. Materials and methods

2.1. Animals and ethics

Experiments were performed on 25–35 g CD1 male mice (Charles River, Gödöllő, Hungary), kept under standard pathogen-free conditions at 24–25 °C and provided with standard rodent chow and water ad libitum. The light/dark cycle was 12 h/12 h. Animal procedures were approved by the local animal ethics committee and National Food Chain Safety Office Animal Health and Animal Welfare Directorate under the number 26/2007/DE MÁB in accordance with the European Communities Council Directives (86/609/ECC) and the Hungarian Act for the Protection of Animals in Research (XXVIII tv. 32§) and compiled with the recommendations of the International Association for the Study of Pain [21] and the Helsinki Declaration. The design of the study was carried out in a manner in which to minimize the number of animals used and their suffering.

2.2. Substances and their application

Mice were doused with 1, 3 or 10 mg/kg sitagliptin or vildagliptin (Nanjing Ange Pharmaceuticals, Nanjing, Jiangsu, China) dissolved in saline by oral gavage (1 ml/100 g). Control groups were given the vehicle in the same amount and way. A single application was used in the case of one-day experiments, while daily application was used in the 21 day long experiments, as suggested by Thomas et al. [22]. Treatments and measurements were implemented 30 min after the oral gavage in every case.

2.3. Allyl-isothiocyanate (AITC)-induced inflammation model

Anesthesia was induced by thiopental (Trapanal, Sandoz, Basle, Switzerland) in an amount of 50 mg/kg intraperitoneally (i.p.), repeated as required. The inner and outer surface of the right ear was then smeared with 1% allyl-isothiocyanate (AITC) (Sigma-Aldrich, Budapest, Hungary) dissolved in paraffin oil, using a cotton-wool stick. This treatment was applied 30 min after the oral gavage (substances dissolved in saline or vehicle in the control group) and the procedure was repeated 45 min after the first application following the instructions of Bánvölgyi [23] and Inoue et al. [24]. Thus the oral administration of gliptins was performed firstly and the induction of inflammation was carried out secondly.

At the end of the experiment the animals were sacrificed by cervical dislocation and ears were stored on -20 °C for the neutrophil accumulation assay.

2.4. Measurement of ear edema

Ear thickness was measured by a micrometer caliper (Oxford Precision, Leicester, England) with 0.1 mm accuracy before the AITC treatment, 15 min after the first AITC application, then by each hour during a 6 hour period after each AITC treatment according to Inoue et al. [24] with slight modifications. Gliptin treatment was performed 30 minutes...
before the commencement of ear edema induction. Data were expressed in micrometers.

2.5. Measurement of neutrophil accumulation

Frozen ear samples were thawed at room temperature, chopped into small pieces, and homogenized in 0.05 M potassium phosphate buffer containing 0.5% HTAB (hexadecyltrimethylammonium bromide, Sigma-Aldrich, Budapest, Hungary), 1 ml buffer/ear. The homogenate was centrifuged at 11,000 g at 4 °C for 10 min and 200 μl of the supernatant was placed into Eppendorf tubes.

Myeloperoxidase activity was assayed by measuring the H2O2-dependent oxidation of 3,3′,5,5′-tetramethylbenzidine (TMB, Sigma-Aldrich, Budapest, Hungary) as suggested by Suzuki et al. [25]. In its oxidized form, TMB has a blue color, which was measured spectrophotometrically at 620 nm. The reaction was performed in 96-well microtiter plates at room temperature. The reaction mixture consisted of 25 μl of the tissue sample, 25 μl of TMB (final concentration 0.16 mM) dissolved in dimethylsulfoxide (DMSO) and 200 μl H2O2 (final concentration 0.24 mM, Sigma-Aldrich, Budapest, Hungary) diluted in 0.08 M phosphate buffer pH 5.4 after Schierwagen et al. [26]. The optical density (OD) was measured at 5 min intervals for 30 min using a microplate reader (FLUOstar OPTIMA, BMG Labtech, Ortenberg, Germany). Data was expressed in arbitrary units of absorbance.

2.6. Induction of arthritis

Chronic arthritis of the right tibiotarsal joint of mice was induced by the subcutaneous injection of 0.1 ml of Freund's complete adjuvant (CFA, killed Mycobacteria suspended in paraffin oil, 1 mg/ml as provided by Sigma-Aldrich, Budapest, Hungary) into the plantar surface of the right hind paw and root of the tail. To enhance systemic effects, an additional injection into the tail was given the following day as described by Helyes et al. [27]. In order to minimize the suffering of mice, short-term general anesthesia was induced by 1% isoflurane (Abbott Laboratories, Budapest, Hungary) delivered in 1:2 oxygen/nitrous oxide mixture.

2.7. Measurement of plasma extravasation in the urinary bladder of mice

Mice were anesthetized by i.p. administration of thiopental (50 mg/kg). A lateral tail vein was cannulated for intravenous administration. 1 or 3 mg of vildagliptin or sitagliptin was administered by oral gavage 30 minutes before the commencement of the capsaicin challenge. Evans blue (30 mg/kg) and 1 minute later capsaicin (1 mg/kg) was injected through the venous cannula. Each animal was sacrificed by transecardiac perfusion with 50 ml of 0.9% w/v saline into the left cardiac ventricle 10 min after intravenous injection of Evans blue at 37 °C. The urinary bladder was then removed and weighed. Excised tissues were incubated in 1 ml of formamide for 48 h and Evans blue content was measured spectrophotometrically at 620 nm and expressed as μg/g wet mass of the tissue.

2.8. Measurement of mechano-nociceptive threshold

Touch sensitivity on the plantar surface was measured with von Frey filaments (Bioseb, Chaville, France) before the experiment, 3, 7, 10, 14, 17, and 21 days following the first CFA administration. The set of 20 monofilaments provided an approximate logarithmic scale of actual force and a linear scale of perceived intensity. Mice were placed into a Plexiglas cage with a pitted floor. Following animal acclimatization the operator placed the monofilament under the animal's paw and pressed against the surface till the animal indicated the pressure sensation by pulling back or shaking its paw, or the monofilament curved without any kind of reaction starting with 0.008 g and ranging up to 300 g.

2.9. Increasing-temperature hot plate test

The plate (Supertech, Pécs, Hungary) in contact with the paws has been slowly warmed up from room temperature and the threshold temperature producing the first nociceptive behavior (e.g., paw licking) was recorded. Since the temperature was increased gradually into the noxious range, stress associated with the testing procedure was minimized. The heated surface dimensions were 110 × 80 mm surrounded by 350 mm high transparent Plexiglas walls. The commanding computer program was set to produce a 3 °C/min temperature increase of the plate as proposed by László et al. [28]. When the hind paw licking or flinching was observed the threshold temperature was recorded. The measurement was terminated at the threshold level or when the plate temperature reached 50 °C to avoid tissue damage [29]. Data were expressed in °C.

2.10. Statistics

Since baseline values of diverse groups were significantly different in all measurements, a baseline correction was carried out on raw values. Baseline corrected values were regarded as primary outcome measures. Two-way ANOVA with replication was used for multiple group analysis with the time point of observation and the treatment option as factors.
for the complete duration of the experiments. Games-Howell tests were used as post hoc analysis for binary comparison of group averages. Significant differences at the 95% confidence interval were recognized, if \( p < 0.05 \). Below 0.001 no numeric values of \( p \) are provided in the text.

### 3. Results

#### 3.1. AITC-induced ear edema

Both orally administered gliptins significantly decreased ear thickness in the complete time period compared to positive control (AITC only) in a dose-dependent manner as seen in Fig. 1. The maximum effect of AITC was measured at 2 hour post-challenge time in either case. For the complete examination period 10 mg sitagliptin as well as 1–10 mg vildagliptin was found to significantly decrease ear edema as compared to positive control.

#### 3.2. AITC-induced neutrophil accumulation

The evolved inflammation was shown by the high level of myeloperoxidase enzyme in the positive control group (AITC only), see Fig. 2. The model is suitable for measuring the extent of inflammation, since these data definitely diverge from the negative control group results. Sitagliptin treatment was found significantly effective in blocking the evolution of inflammation; every examined dose could reverse inflammation. (Blind samples were not included in the hypothesis testing.) The effect of vildagliptin treatment was similar to that of sitagliptin, although the dose of 3 mg/kg had only an insignificant impact.

#### 3.3. Measurement of plasma extravasation in the urinary bladder of mice

The capsaicin-induced plasma extravasation in urinary bladders of mice was inhibited by sitagliptin (1 mg \( p = 0.025 \) and 3 mg \( p < 0.001 \)) and vildagliptin (both 1 mg and 3 mg \( p < 0.001 \)) significantly (Fig. 4). Difference in action was seen between the higher doses (3 mg/kg) of vildagliptin and sitagliptin. The lower dose of sitagliptin (1 mg/kg) produced the least significant inhibition compared to the control.

#### 3.4. Touch sensitivity in CFA-induced arthritis

Results show that the mechano-nociceptive threshold of the untreated group was significantly higher than in the CFA treated (positive

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control) group during the whole 21 day experimental period (Fig. 3). In the gliptin treated groups every threshold was significantly lower than in the negative control group; consequently neither sitagliptin nor vildagliptin treatment was able to hinder the evolution of allodynia. Sitagliptin was significantly harmful in an amount of 3 and 10 mg/kg doses as compared to the positive control, meanwhile vildagliptin was able to hinder the evolution of allodynia. Further, the GLP-1 receptor (GLP-1R) is expressed in lymphoid tissue and the numbers of CD4+ and CD8+ T-cells in lymph nodes was shown to increase after exenatide (a GLP-1R agonist) treatment. It could also reduce the number of CD4+ CD25+ Foxp3+ regulatory T-cells in the thymus, but not in the spleen [31], thus playing a regulatory role in the immune system and can influence inflammatory processes [32]. However, Kim et al. [33] were unable to detect the effect of either GIP or GLP-1 on splenic or thymic CD4+ T-cell migration in vitro [33]. Eosinophil cell recruitment (in allergic asthma or in atopic dermatitis) is described to be mediated by CCL11 (eosinophil chemotactic protein) and the recruitment proved to be more effective after pharmacological inhibition of DPP-4 enzyme or in DPP-4-deficient F344 rats [34]. The activation of transient receptor potential ankyrin 1 (TRPA1) evokes nociception through substance P release from the primary sensory neurons; p38 mitogen-activated protein kinase (p38 MAPK) inhibitor SB203580 significantly attenuated AITC-evoked substance P release [35]. Allcyloisothiocyanate is capable of inducing ear edema in the proper dose as described earlier [23]; the maximum auricle swelling was measured in the second hour. Both examined chemicals, sitagliptin and vildagliptin were able to decrease the AITC-induced inflammation in a dose-dependent manner however, sitagliptin had a higher impact. This effect cannot be explained by the regulatory role of GLP-1 on p38 MAPK, as it was described as an inducer [36]; neither can it be attributed to the effect of gliptins on substance P metabolism [37]. Moreover, the physiological role of GLP-1 is so dominant that its inhibition can still override the p38 MAPK-inhibitor property and the algogenic effect of elevated substance P. Treatment by DPP4 inhibitor 40 significantly reduced the severity of experimental allergic encephalomyelitis (EAE), in mice conceivably through up-regulating TGF-beta 1 [18]. Furthermore, a dose-dependent inhibition of the secretion of the pro-inflammatory cytokine TNF-alpha was measured in vitro [18]. The ability of gliptins to

4. Discussion

According to our present data, we can conclude that the studied gliptins had a dose-dependent anti-inflammatory effect in in vivo mouse models. The applied methods were sensitive enough to detect the action of gliptins. Dipeptidyl peptidase inhibitors were reviewed as an emerging drug class for various inflammatory diseases [7]. The anti-inflammatory action of these drugs were described in human studies [8] and for exenatide [19]. Sitagliptin significantly improves endothelial function and inflammatory state in patients with coronary artery disease and uncontrolled diabetes mellitus [30], forming a milestone in the way towards widening the spectrum of gliptins’ indication. Moreover, the GLP-1 receptor (GLP-1R) is expressed in lymphoid tissue and the numbers of CD4+ and CD8+ T-cells in lymph nodes was shown to increase after exenatide (a GLP-1R agonist) treatment. It could also reduce the number of CD4+ CD25+ Foxp3+ regulatory T-cells in the thymus, but not in the spleen [31], thus playing a regulatory role in the immune system and can influence inflammatory processes [32]. However, Kim et al. [33] were unable to detect the effect of either GIP or GLP-1 on splenic or thymic CD4+ T-cell migration in vitro [33]. Eosinophil cell recruitment (in allergic asthma or in atopic dermatitis) is described to be mediated by CCL11 (eosinophil chemotactic protein) and the recruitment proved to be more effective after pharmacological inhibition of DPP-4 enzyme or in DPP-4-deficient F344 rats [34]. The activation of transient receptor potential ankyrin 1 (TRPA1) evokes nociception through substance P release from the primary sensory neurons; p38 mitogen-activated protein kinase (p38 MAPK) inhibitor SB203580 significantly attenuated AITC-evoked substance P release [35]. Allyl-isothiocyanate is capable of inducing ear edema in the proper dose as described earlier [23]; the maximum auricle swelling was measured in the second hour. Both examined chemicals, sitagliptin and vildagliptin were able to decrease the AITC-induced inflammation in a dose-dependent manner however, sitagliptin had a higher impact. This effect cannot be explained by the regulatory role of GLP-1 on p38 MAPK, as it was described as an inducer [36]; neither can it be attributed to the effect of gliptins on substance P metabolism [37]. Moreover, the physiological role of GLP-1 is so dominant that its inhibition can still override the p38 MAPK-inhibitor property and the algogenic effect of elevated substance P. Treatment by DPP4 inhibitor 40 significantly reduced the severity of experimental allergic encephalomyelitis (EAE), in mice conceivably through up-regulating TGF-beta 1 [18]. Furthermore, a dose-dependent inhibition of the secretion of the pro-inflammatory cytokine TNF-alpha was measured in vitro [18]. The ability of gliptins to

Fig. 4. Inhibition of capsaicin-induced plasmaextravasation in urinary bladders of mice by Vildagliptin and Sitagliptin. Gliptins were administered by oral gavage in 1 or 3 mg/kg dose 30 minutes before the capsaicin (1 mg/kg) intravenous challenge. Evans blue dye was administered in 30 mg/kg i.v. and the plasmaextravasation was determined spectrophotometrically at 620 nm wave length. Error bars denote standard error of the mean. * and × denote significant differences to negative control and to vildagliptin 1 mg, respectively as assessed by Games-Howell post hoc test.

3.5. Increasing-temperature hot plate test in CFA-induced arthritis

Threshold temperature of the untreated group was significantly lower than in the control group during the whole 21 day experimental period (Fig. 3). In the gliptin treated groups every threshold was significantly lower than in the negative control group; consequently neither sitagliptin nor vildagliptin treatment was able to hinder the evolution of allodynia. Sitagliptin was significantly harmful in an amount of 3 and 10 mg/kg doses as compared to the positive control, meanwhile vildagliptin remained ineffective in all doses.

Fig. 5. Time evolution of the thermo-nociceptive threshold in Freund’s complete adjuvant (CFA)-induced arthritis model in mice as a function of the amount of A) sitagliptin or B) vildagliptin administered by oral gavage. Error bars denote standard error of the mean. Lines between markers guide the eye only. For the complete time period * (p < 0.001), × (p < 0.05), and # (p < 0.05) showed significant differences to negative, positive control, and 1 mg sitagliptin, and # (p < 0.001), × (p < 0.05), * (p < 0.05), and $ (p < 0.01) to negative, positive control, 1 mg, and 3 mg vildagliptin, respectively as assessed by Games-Howell post hoc test.
regulate TNF-α, INF-gamma, and a variety of interleukins can be attributed to the DPP4 inhibitory activity, because the compounds used in the present series of experiments have a high specificity to DPP4 and probably do not have any inhibitory effect on DPP8 or 9 in the applied doses [38]. However, vildagliptin ineffectiveness in two models (myeloperoxidase measurement and touch sensitivity in CFA-induced arthritis) at 3 mg/kg can be the result of DPP-9 activity attenuation having 66 nM IC50 value in vitro compared to 130 nM IC50 for sitagliptin [39]. Inhibition of DPP-8/9 can lead to the development of adverse effects in rodents [9,40], but other studies state that the inhibition of DPP-8/9 do not have any clinical consequence [41]. A reduced expression of nitrosative stress and inflammation hallmarks within the brain of chronically administered sitagliptin was described earlier in a mouse model of Alzheimer’s disease [42]. An explanation could easily arise considering the fact that GLP-1 can have growth-factor-like properties similar to insulin and the anti-inflammatory activity is a secondary action [43]. In our experiments, the anti-inflammatory action seems to be direct as demonstrated by the accumulated number of neutrophil cells (measured by myeloperoxidase enzyme activity) in the inflamed ear; this accumulation could be inhibited by the glititin pre-treatment. Similarly to the above mentioned tests, sitagliptin treatment had a higher impact in the compensation of the CFA-induced arthritis, where vildagliptin showed no effectiveness. In case of measuring the high temperature sensitivity, both substances showed equal effectiveness. Our results lead to the conclusion that sitagliptin has a stronger influence on the evolution of inflammation; however, vildagliptin showed higher effectiveness in the inhibition of capsaicin-induced plasma extravasation in the urinary bladder. Although the investigated molecules have the same effectiveness in the treatment of type-2 diabetes, it seems that they do not act in the same way, in the same immune response. Both substances represent promising options for the therapy of inflammatory disorders.

Conflict of interest

The authors declare that there are no conflicts of interest.

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