Abstract

Strawberry (Fragaria ananassa) is one of the richest sources containing a wide variety of nutritive compounds. Anti-inflammatory activities of fermented rice cake made of strawberry powder as well as rice powder were evaluated. The fermented rice cake containing strawberry powder (SRC) significantly and dose-dependently inhibited NO production in LPS-stimulated RAW264.7 cells without cytotoxicity. Also, SRC effectively suppressed inflammatory gene expression, including iNOS, COX-2, IL-1β, IL-6, and TNF-α. In addition, the production of PGE₂, IL-1β, IL-6, and TNF-α was significantly reduced. Furthermore, the anti-inflammatory effect of SRC was investigated using carrageenan-induced paw edema of ICR mice. It was demonstrated that pre-orally administration of SRC at a dose of 50 and 100 mg/kg BW significantly inhibited paw edema induced by carrageenan. This study suggested that the anti-inflammation activities of strawberry rice cake give the potential for increasing the commercialization of rice cake and rice products.

Introduction

Rice cakes are the popular traditional foods in Asian countries such as Korea and China [1]. They are mainly made with rice flour or other grains and they are prepared by grinding, streaming, boiling, or frying from the different ingredients and different manufacturing process [2–4]. Many studies have developed functional products by adding the powder of several ingredients into rice cakes such as mulberry, sweet potato, and ginseng [5–7]. Some powders such as almond, maqui berry, pumpkin, and chickpea [8–11], have been reported to exhibit antioxidant effects, while the anti-inflammatory effects in fermented rice cake by adding some ingredients have not been extensively studied.

Inflammation is a complex mechanism of interactions among soluble immune factors and related cells that can occur in any tissue in response to traumatic, infectious, post-ischemic, toxic, or autoimmune [12]. It is a protective biological response to harmful stimulation,
pathogens, or irritants in vascular tissues that attempts to eliminate infectious stimulation [13]. Macrophages are considered to play essential roles in inflammation [14], and interleukin-1β (IL-1β), IL-6, and tumor necrosis factor-α (TNF-α) are produced by activated macrophages, which are the important mediators of the inflammatory response, and are involved in a variety of cellular activities, including cell proliferation, differentiation, and apoptosis [15, 16].

Strawberry (Fragaria ananassa) contains the bioactive compounds of phenolic compounds, vitamins, minerals, polysaccharides, that there have many biological activities [17]. Lin and Tang reported that strawberries showed immunomodulatory activity by stimulated splenocyte proliferation from BALB/c mice [18]. In addition, strawberry juice exhibited the anti-inflammatory effect on murine peritoneal macrophage [19] and also inhibited ROS and NO production in LPS-stimulated RAW264.7 macrophage cells by strawberry extracts [20]. The preliminary research study showed that a mixture of rice powder and strawberry powder containing a ratio of 10: 90 effectively inhibits inflammatory responses [21]. In the present study, the anti-inflammatory properties of a rice cake made from a mixture of strawberry and rice powder (10: 90) were investigated in vitro macrophage and in vivo mouse models.

Materials and methods

Fermented strawberry rice cake sample preparation

Fermented strawberry rice cake (SRC), L-glutamine rice cake (LRC), and rice cake (RC) were provided from Sanghwa F&B Inc. (Gangneung, Korea) after manufacturing. Briefly, SRC and LRC were made from the mixture of rice cake (90%) and strawberry powder or L-glutamine (10%) as described in our previous report [21]. SRC, LRC, and RC were refrigerated for 24 h and then they were lyophilized using a freeze dryer. The freeze-dried rice cake was ground in a blender and was collected as power for further experiments. All samples (SRC, LRC, RC, strawberry powder, and L-glutamine) were dissolved with deionized sterile before use.

Animal cell culture and sample treatments

Rodent macrophages, RAW264.7 cells were obtained from Koran Cell Line Bank (KCLB, Cat# 40071, RRID: CVCL_0493). The cells were maintained at 37˚C in a humidified incubation with 5% CO₂ in RPMI (Gibco™, Waltham, USA, Cat# 11875–093) supplemented with 10% fetal bovine serum (FBS, Welgene, Korea, Cat# S001-07) and 1% streptomycin/ampicillin (Welgene, Korea, Cat# LS202-02). RAW264.7 cells (1×10⁵ cells/ well) were treated with the various concentrations of SRC or LRC (0.78, 1.56, 3.12, and 6.25 mg/mL), with the control; strawberry powder (STP; 0.625 mg/mL), RC (5.625 mg/mL), and L-glutamine (Gln; 0.625 mg/mL) for 1 h. After that, the cells were stimulated with 1 μg/mL of lipopolysaccharides (LPS from Escherichia coli O111:B4, Sigma-Aldrich, USA, Cat# L4391-1MG) and incubated for another 24 h.

Measurement of cell proliferation and NO production

After incubation 24 h, the cultured medium and Griess reagent (Promega, WI, USA, Cat# G2930) was used for the evaluation of NO production [22]. The cell proliferation was analyzed using EZ-Cytox Cell Viability Assay Kit Kit (DaeilLab Service, Seoul, Korea, Cat# EZ-3000) as described by Kim et al. [23].

The cellular proliferation ratio (%) was calculated based on the following formula:

\[
\text{Macrophage proliferation ratio} \% = \frac{\text{the absorbance of the test group}}{\text{the absorbance of the control group}} \times 100
\]

Competing interests: The authors have declared that no competing interests exist.
Analysis of mRNA expression by quantitative real-time PCR

The cells were extracted from the total RNA using Tri reagent® (Molecular Research Center, Cincinnati, OH, USA, Cat# TR118). The total RNA was synthesized to the first-strand cDNA by the High-capacity cDNA reverse transcription kit (Applied Biosystems, Foster City, CA, USA, Cat# 4368814), according to the manufacturer’s instructions. Real-time PCR was subsequently performed using the QuantStudio™ 3 FlexReal Time PCR System (Applied Biosystems, Foster City, CA, USA) and TB Green™ Premix Ex Taq™ II (Takara Bio Inc., Shiga, Japan, Cat# RR820A). These reactions were conducted using the primers specific to the target gene of iNOS, COX-2, IL-1β, IL-6, TNF-α, and β-actin, that the sequences were summarized in Table 1.

Measurement of PGE₂, TNF-α, IL-1β and IL-6

After treatment, the supernatants were collected and centrifuged at 1000 x g for 20 min. In accordance with the manufacturer’s instructions, the concentrations of PGE₂, IL-1β, IL-6, and TNF-α were determined by the PGE₂ ELISA kit (Enzo Life Sciences, Inc. USA, Cat# ADI-900-001), IL-1β (Abcam, USA, Cat# ab197742), IL-6 (Abcam, USA, Cat# ab100712), and TNFα (Abcam, USA, Cat# ab208348), respectively.

Animals

Male ICR mice with 28 ± 2 g of body weight (BW) were purchased from Orient Bio Inc. (Seongnam, Korea). The animals were kept under controlled conditions with a standard laboratory diet and water for one week before starting the experiment. These experimental protocols were approved by the Institutional Animal Care and Use Committee (IACUC) of Gangneung-Wonju National University, Korea (Approval Number: GWNU-2018-20).

Carrageenan-induced paw edema in ICR mice

To determine the anti-inflammatory effects in an animal model, carrageenan-induced paw edema was investigated [24, 25]. The animals were randomly divided into six groups (5 mice for each group). Group A received saline solution as control, Group B received STP at 10 mg/kg BW, Group C received RC at 100 mg/kg BW, Group D received LRC at 100 mg/kg BW, Group E-F received SRC at the dose of 50 and 100 mg/kg BW. All treatments were administered orally in mice. After 1 h of oral administration, the suspension of carrageenan at 0.5 mg/25 μL (Sigma-Aldrich, USA, Cat# C1867-1G) was injected into the subplantar tissue of the right hind paw, while the left hind paw was injected with saline solution. Paw edema was measured after carrageenan injection at 90, 180, 270, and 360 min to assess the difference in footpad thickness between the right and left foot.
Statistical analysis
Data were analyzed by One-way ANOVA (Holm–Sidak post hoc multiple comparison test) under the software of 'Statistix 8.1' Statistics (Tallahassee, FL, USA). The statistical differences were considered significant at $p < 0.05$.

Results
Effects of SRC and LRC on LPS-induced cell cytotoxicity and NO production in RAW264.7 cells
The cytotoxicity of SRC and LRC was determined using RAW264.7 cells with the treatment of different concentrations of rice cake samples as well as negative and positive controls. Cellular proliferation of SRC and LRC treated cells was shown in Fig 1A, in which any samples did not provide any toxicity to RAW264.7 cells.

In addition, the anti-inflammatory effects of SRC and LRC were analyzed using the production of NO production as an important factor for inflammation [26]. Fig 1B shows that LPS led to inflammation by stimulating NO production compared to the normal RAW264.7 cells. The treatment of SRC and LRC significantly inhibited LPS-induced NO production according to the SRC and LRC concentration. Moreover, the treatment with 6.25 mg/mL of SRC and LRC gave lower NO production compared to the normal rice cake group.

Effects of SRC and LRC on LPS-induced mRNA expressions of immune-associated genes in RAW264.7 cells
As shown in Fig 2, iNOS and COX-2 expressions, critical inflammation-associated genes were concentration-dependently inhibited in LPS-induced and SRC and LRC treated macrophage cells. Similar to the mRNA expression of IL-1β, IL-6, and TNF-α which are the pro-inflammatory cytokines [14], were also dose-dependently decreased (Fig 2). In addition, both SRC and LRC treatments at 6.25 mg/mL exhibited the highest anti-inflammatory effect, when compared with the strawberry powder, L-glutamine, and rice cake treatments.

Fig 1. Effect of SRC and LRC on LPS-stimulated RAW264.7 macrophage cells. (A) Macrophage proliferation. (B) NO production. The results are presented as the mean ± SD ($n = 3$). Significant differences are $p < 0.05$ compared with RPMI. SRC = strawberry rice cake, LRC = L-glutamine rice cake, STP = strawberry powder, L-glutamine = Gln and RC = rice cake.

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Effects of SRC and LRC on LPS-induced production of PGE$_2$, IL-1$\beta$, IL-6, and TNF-$\alpha$

In order to evaluate the anti-inflammatory effects of SRC and LRC on LPS-stimulated RAW264.7 cells, the concentrations of pro-inflammatory cytokines and PGE$_2$ were measured.
using an ELISA. As shown in Fig 3, the levels of PGE₂, IL-1β, IL-6, and TNF-α were significantly increased by the LPS treatment. Furthermore, treatment with both SRC and LRC reduced levels of all four in a dose-dependent manner.

**Effect of SRC on carrageenan-induced paw edema**

The present study was carried out to evaluate the anti-inflammatory effect of SRC in a model of inflammation using the carrageenan-induced inflammation mice system. Fig 4 and Table 2 showed that oral administration of SRC at the dose of 50 and 100 mg/kg BW reduced the size of paw edema induced by carrageenan. After carrageenan injection, the size difference between the left and right paw was gradually increased over a period of 6 h in the control group (received saline). In contrast to SRC administration, the difference between left and right paw reached the maximum at 2 h after the induction of carrageenan, and then gradually decreased. Moreover, the administration of strawberry and rice cake reduced the difference between left and right paw after 4.5 h of carrageenan induction.

**Discussion**

Inflammation is a defense and prevention against harmful stimuli. The immune response to bacterial infections is induced by LPS by triggering a variety of intracellular signaling events
Macrophages are mainly involved in acute and chronic inflammatory responses which produce NO to enhance to eliminate microorganisms or to regulate inflammation [28, 29], however, excessive production of NO is considered toxic to the host tissue [30]. Various compounds from plants have been reported to play potential roles in many pharmacological properties including anti-inflammatory, anti-cancer, antioxidant, cardiovascular, and neurological diseases [31–33].

Strawberry is reported to have an anti-inflammatory effect via reducing the production of NO and inhibiting the inflammatory mediators and cytokines in LPS-stimulated RAW264.7 cells [34–37]. Strawberry extract exhibited anti-inflammatory effects, reducing the NO and ROS intracellular production as well as inflammatory markers (TNF-α, IL-1β, IL-6 and, IL-10) through the activation of the Nrf2 pathway, and NF-κB signaling pathway in vitro [27].

Table 2. The different size between left and right paw (mm).

| Group | Treatment | Dose (mg/kg BW) | Time after carrageenan-induced (min) | 90  | 180 | 270  | 360 |
|-------|-----------|----------------|-------------------------------------|-----|-----|------|-----|
| A     | Control   | -              | 0.73±0.012                          | 1.10±0.044 | 1.34±0.031 | 1.61±0.067 |
| B     | STP       | 10             | 0.73±0.006                          | 1.00±0.044 | 1.18±0.047 | 0.95±0.015 |
| C     | RC        | 100            | 0.79±0.015                          | 1.04±0.010 | 1.26±0.030 | 1.13±0.031 |
| D     | LRC       | 100            | 0.36±0.035                          | 0.74±0.025 | 0.90±0.031 | 1.05±0.015 |
| E     | SRC       | 50             | 0.95±0.053                          | 1.08±0.087 | 0.93±0.021 | 0.65±0.031 |
| F     | SRC       | 100            | 0.77±0.015                          | 0.94±0.010 | 0.82±0.025 | 0.63±0.030 |

*p < 0.05 when compared with control group. Results represent the mean ± SD of 5 animals for each group (n = 5).
Stimulation [20]. Strawberry also significantly reduced the inflammation-associated biomarkers in the clinical study [32, 38]. Similarly, serum collected from rodents fed blueberry and strawberry-enriched diets showed anti-inflammatory activity [39]. Previous research examined the anti-inflammatory activities of a strawberry-rice powder mixture as a material of fermented rice cake on RAW264.7 cells induced by LPS and mouse models induced by carrageenan [21]. Nevertheless, no research has been reported on the anti-inflammatory effects of rice cake supplemented with strawberry powders, so our experiments used LPS-stimulated RAW264.7 cells and carrageenan-induced inflammation mice to investigate the anti-inflammatory properties of rice cake supplemented with strawberry powders.

The current study showed that the supplementation with strawberry powders in fermented rice cake exhibited anti-inflammatory activity. NO production, critical immune-regulatory biomarker for inflammation [40], was significantly decreased according to SRC concentration (Fig 1B). The pro-inflammatory cytokines such as IL-1β, IL-6, and TNF-α have been known to regulate immune systems in macrophages [15, 16]. The current study also showed that the expression of IL-1β, IL-6, and TNF-α was significantly and dose-dependently decreased in LPS-stimulated RAW264.7 cells when the cells were pre-treated with SRC (Fig 2). Among the fermented products, red mold rice by Monascus purpureus, was reported to inhibit NO production in LPS-stimulated RAW264.7 cells [41], and the pelargonidin-3-O-glucoside, ellagic acid, and polyphenolic extracts, which were isolated from strawberry also inhibited the production of NO, TNF-α, and IL-6 and the expression of pro-inflammatory cytokines [34–37]. Moreover, our results showed COX-2 which is a key mediator of inflammatory pathway [42] also significantly inhibited in LPS-stimulated RAW264.7 cells. Similarly, the LPS-induced COX-2 expression was decreased by the polyphenolic extracts from strawberry [37].

Injection of carrageenan into the mouse paw leads to local inflammation which is a suitable method for evaluating anti-inflammatory agents [43]. Carrageenan-induced rat paw edema is a widely used test to determine the anti-inflammatory activity [44–46]. Mouse paw edema has been increasingly used to test new anti-inflammatory drugs as well as to study the mechanisms involved in inflammation [45]. Our results showed that the anti-inflammatory effect was observed after carrageenan-induced inflammation in mice that received SRC via reducing the size difference between the left and right paw of SRC-treated mice. These results are similar to some reports which showed the anti-inflammatory activity of Berberidaceae roots extracts as well as Berberis root and bark extract on carrageenan-induced mice paw [25, 47–49]. However, any reports which studied fermented rice cake containing anti-inflammatory effects have not been found.

Conclusions

Our results demonstrated that SRC exerts anti-inflammatory effects both in vitro and in vivo physiological systems. SRC inhibited the LPS-induced NO production and pro-inflammatory cytokines on RAW264.7 macrophages cells. Furthermore, SRC also suppressed the paw edema thickness on the carrageenan-induced mice model. Therefore, these results suggested that fermented rice cake using strawberry powder has a potential traditional food to provide anti-inflammatory effects under several disease conditions as a supplementary diet.

Author Contributions

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References
1. Wang J, Park JH, Choi NJ, HA S, Oh DH. Microbiological analysis of rice cake processing in Korea. J Food Prot. 2016; 79(1): 157–162. https://doi.org/10.4315/0362-028X.JFP-15-237 PMID: 26735044.
2. Kim DH, Lim YT, Park YJ, Yeon SJ, Jang KI. Antioxidant activities and physicochemical properties of tteokbokki rice cakes containing cinnamon powder. Food Sci Biotechnol. 2014; 23(2): 425–430. https://doi.org/10.1007/s10068-014-0058-8
3. Meng LW, Kim SM. Effects of different carbohydrates on the physicochemical properties of rice flour, and the quality characteristics of fermented rice cake. Food Sci Biotechnol. 2020; 29(4): 503–512. https://doi.org/10.1007/s10068-019-00693-7 PMID: 32296561.
4. Wang H, Wang M, Chen J, Tang Y, Dou J, Yu J, et al. A polysaccharide from Strongylocentrotus nudus eggs protects against myelosuppression and immunosuppression in cyclophosphamide-treated mice. Int Immunopharmacol. 2011; 11(11): 1946–1953. https://doi.org/10.1016/j.intimp.2011.06.006 PMID: 21723424.
5. Kang HJ, Kim S, Kum JS, Lim JK. Effect of ginseng powder on quality characteristics of instant rice cake (Baekseolgi). J Korean Soc Food Sci Nutr. 2010; 39(3): 435–442. https://doi.org/10.3746/jkfn.2010.39.3.435
6. Lee J, Kim B. Effect of added sweet potato flour on the quality characteristics of the Korean traditional steamed rice cake, Backsulki. Food Eng Prog. 2010; 14(2): 135–145.
7. Park GY, Liu Q, Hong JS, Chung HJ. Anti-staling and quality characteristics of Korean rice cake affected by mulberry (Morus alba L.) leaf powder fortification. J Cereal Sci. 2021; 97: 103133. https://doi.org/10.1016/j.jcs.2020.103133
8. Cho N, Chung HJ. Quality characteristics and antioxidant activity of Sulgidduk added with maquiberry powder. Korean J Food Preserv. 2016; 23(7): 945–952. https://doi.org/10.11002/kjfp.2016.23.7.945
9. Park GH, Ju HM, Park YJ, Lee EH, Kim HS, Ma RN, et al. Antioxidant activity of Korean rice cake added Cicer arietinum for post-menopausal women. Pak J Nutr. 2015; 14(5): 686–692. https://doi.org/10.3923/pjn.2015.686.692
10. Song KY, Zhang Y, Joung KY, Kim YS. Effects of pumpkin (Cucurbita moschata Duch.) leaf powder on quality characteristics, antioxidant activities, and retarding retrogradation by shelf-life of Sulgidduk (rice cake). J Korean Soc Food Sci Nutr. 2016; 45(12): 1792–1798. https://doi.org/10.3746/jkfn.2016.45.12.1792
11. Yu HN, Song JH, Kim MR. Quality characteristics and antioxidant activities of Sulgidduk added with almond powder. J Korean Soc Food Sci Nutr. 2017; 46(7): 809–815. https://doi.org/10.3746/jkfn.2017.46.7.809
12. Nathan C. Points of control in inflammation. Nature. 2002; 420(6917): 846–852. https://doi.org/10.1038/nature01320 PMID: 12490957.
13. Lee E, Shin SY, Kim JK, Woo ER, Kim YM. Anti-inflammatory effects of amentoflavone on modulation of signal pathways in LPS-stimulated RAW264.7 cells. Bull Korean Chem Soc. 2012; 33(9): 2878–2882. https://doi.org/10.5012/bkcs.2012.33.9.2878
14. Nathan C. Nitric oxide as a secretory product of mammalian cells. FASEB J. 1992; 6(12): 3051–3064. https://doi.org/10.1096/fasebj.6.12.1381691 PMID: 1381691.

15. Yencilek F, Yildirim A, Yılmaz SG, Altinkılıç EM, Dalan AB, Bastug Y, et al. Investigation of interleukin-1β polymorphisms in prostate cancer. Anticancer Res. 2015; 35(11): 6057–6061. PMID: 26504029.

16. Gopinath VK, Musa M, Samsudin AR, Kosresno W. Role of interleukin-1β and tumour necrosis factor-α on hydroxyapatite-induced phagocytosis by murine macrophages (RAW264.7 cells). Br J Biomed Sci. 2006; 63(4): 176–178. https://doi.org/10.1080/09674845.2006.11978094 PMID: 17201208.

17. Giampieri F, Alvarez-Suarez JM, Mazzoni L, Romandini S, Bompadre S, Diamanti J, et al. The potential impact of strawberry on human health. Nat Prod Res. 2013; 27(4–5): 448–455. https://doi.org/10.1080/14786419.2012.706294 PMID: 22788743.

18. Lin JY, Tang CY. Determination of total phenolic and flavonoid contents in selected fruits and vegetables, as well as their stimulatory effects on mouse splenocyte proliferation. Food Chem. 2007; 101(1): 140–147. https://doi.org/10.1016/j.foodchem.2006.01.014.

19. Lin JY, Tang CY. Strawberry, loquat, mulberry, and bitter melon juices exhibit prophylactic effects on LPS-induced inflammation using murine peritoneal macrophages. Food Chem. 2008; 107(4): 1587–1596. https://doi.org/10.1016/j.foodchem.2007.10.025.

20. Gasparrini M, Forbes-Hernandez TY, Giampieri F, Afrin S, Alvarez-Suarez JM, Mazzoni L, et al. Anti-inflammatory effect of strawberry extract against LPS-induced stress in RAW264.7 macrophages. Food Chem Toxicol. 2017; 102: 1–10. https://doi.org/10.1016/j.fct.2017.01.018 PMID: 28130090.

21. Monnai C, Nam JH, Lim JH, Rod-in W, Lee TH, Park WJ. Anti-inflammatory activities of the mixture of strawberry and rice powder as materials of fermented rice cake on RAW264.7 macrophage cells and mouse models. Food Sci Biotechnol. 2021; 30(11): 1409–1416. https://doi.org/10.1007/s10068-021-00929-5 PMID: 34790424.

22. Cao RA, Lee Y, You S. Water soluble sulfated-fucans with immune-enhancing properties from Ecklonia cava. Int J Biol Macromol. 2014; 67: 303–311. https://doi.org/10.1016/j.ijbiomac.2014.03.019 PMID: 24661888.

23. Kim JB, Han AR, Park EY, Kim JY, Cho W, Lee J, et al. Inhibition of LPS-induced iNOS, COX-2 and cytokines expression by poncirin through the NF-κB inactivation in RAW264.7 macrophages. Biol Pharm Bull. 2007; 30(12): 2345–2351. https://doi.org/10.1248/bpb.30.2345 PMID: 18057724.

24. Lee CJ, Chen LG, Liang WL, Wang CC. Anti-inflammatory effects of Punica granatum Linne in vitro and in vivo. Food Chem. 2010; 118(2): 315–322. https://doi.org/10.1016/j.foodchem.2009.04.123.

25. Yesilada E, Küpeli E. Berberis crataegina DC. root exhibits potent anti-inflammatory, analgesic and febrifuge effects in mice and rats. J Ethnopharmacol. 2002; 79(2): 235–248. https://doi.org/10.1016/s0378-8741(02)00092-5 PMID: 11801387.

26. Lechner M, Lirk P, Rieder J. Inducible nitric oxide synthase (iNOS) in tumor biology: the two sides of the same coin. Semin Cancer Biol. 2005; 15(4): 277–289. https://doi.org/10.1016/j.semcancer.2005.04.004 PMID: 15914026.

27. Palma A, Jarrah AS, Tieri P, Cesareni G, Castiglione F. Gene regulatory network modeling of macrophage differentiation corroborates the continuum hypothesis of polarization states. Front Physiol. 2018; 9: 1659–1659. https://doi.org/10.3389/fphys.2018.01659 PMID: 30546316.

28. Korhonen R, Lahti A, Kankaanranta H, Moilanen E. Nitric oxide production and signaling in inflammation. Curr Drug Targets Inflamm Allergy. 2005; 4(4): 471–479. https://doi.org/10.2174/156801005526359 PMID: 16101524.

29. Fujiwara N, Kobayashi K. Macrophages in inflammation. Curr Drug Targets Inflamm Allergy. 2005; 4(3): 281–286. https://doi.org/10.2174/156801005526359 PMID: 16101534.

30. Hussain QA, McKay LJ, Gonzales-Marín C, Allaker RP. Detection of adrenomedullin and nitric oxide in different forms of periodontal disease. J Periodontal Res. 2016; 51(1): 16–25. https://doi.org/10.1111/jre.12273 PMID: 25866935.

31. Ghosemian M, Owlia S, Owlia MB. Review of anti-inflammatory herbal medicines. Adv Pharmacol Sci. 2016; 2016: 9130979–9130979. https://doi.org/10.1155/2016/9130979 PMID: 27247570.

32. Giampieri F, Forbes-Hernandez TY, Gasparrini M, Alvarez-Suarez JM, Afrin S, Bompadre S, et al. Strawberry as a health promoter: an evidence based review. Food Funct. 2015; 6(5): 1386–1398. https://doi.org/10.1039/c5fo00147a PMID: 25803191.

33. Recio M, Andujar I, Ríos J. Anti-inflammatory agents from plants: progress and potential. Curr Med Chem. 2012; 19: 2088–2103. https://doi.org/10.2174/092986712800229069 PMID: 22414101.

34. Duarte LJ, Chaves VC, Nascimento MVPdS, Calvete E, Li M, Ciraolo E, et al. Molecular mechanism of action of Pelargonidin-3-O-glucoside, the main anthocyanin responsible for the anti-inflammatory effect of strawberry fruits. Food Chem. 2018; 247: 56–65. https://doi.org/10.1016/j.foodchem.2017.12.015 PMID: 29277228.
35. Gu I, Brownmiller C, Stebbins NB, Mauroumoustakos A, Howard L, Lee S-O. Berry phenolic and volatile extracts inhibit pro-inflammatory cytokine secretion in LPS-stimulated RAW264.7 cells through suppression of NF-κB signaling pathway. Antioxidants. 2020; 9(9). https://doi.org/10.3390/antioxid9090871 PMID: 32942640.

36. Lee J, Kim S, Namgung H, Jo YH, Bao C, Choi HK, et al. Ellagic acid identified through metabolomic analysis is an active metabolite in strawberry (‘Seolhyang’) regulating lipopolysaccharide-induced inflammation. J Agric Food Chem. 2014; 62(18): 3954–3962. https://doi.org/10.1021/jf4038503 PMID: 24195637.

37. Van de Velde F, Esposito D, Grace MH, Pirovani ME, Lila MA. Anti-inflammatory and wound healing properties of polyphenolic extracts from strawberry and blackberry fruits. Food Res Int. 2019; 121: 453–462. https://doi.org/10.1016/j.foodres.2018.11.059 PMID: 31108769.

38. Schell J, Scofield RH, Barrett JR, Kurien BT, Betts N, Lyons TJ, et al. Strawberries improve pain and inflammation in obese adults with radiographic evidence of knee osteoarthritis. Nutrients. 2017; 9(9): 949–961. https://doi.org/10.3390/nu9090949 PMID: 28846633.

39. Rutledge GA, Fisher Fau—Miller MG Dr, Miller Mg Fau—Kelly ME, Kelly Me Fau—Bielsinski DF, Bielsinski Di Fau—Shukitt-Hale B, Shukitt-Hale B. The effects of blueberry and strawberry serum metabolites on age-related oxidative and inflammatory signaling in vitro. Food Funct. 2019; 10(12). https://doi.org/10.1039/c9fo01913h PMID: 31746877.

40. Hu SS, Bradshaw HB, Chen JS, Tan B, Walker JM. Prostaglandin E2 glycerol ester, an endogenous COX-2 metabolite of 2-arachidonoylglycerol, induces hyperalgesia and modulates NFκB activity. Br J Pharmacol. 2008; 153(7): 1538–1549. https://doi.org/10.1038/bjp.2008.109 PMID: 18297109.

41. Hsu YW, Hsu LC, Chang CL, Liang YH, Kuo YH, Pan TM. New anti-inflammatory and anti-proliferative constituents from fermented red mold rice Monascus purpureus NTU 568. Molecules. 2010; 15(11). https://doi.org/10.3390/molecules15117815 PMID: 21060290.

42. Gandhi J, Khera L, Gaur N, Paul C, Kaul R. Role of modulator of inflammation cyclooxygenase-2 in gammaherpesvirus mediated tumorigenesis. Front Microbiol. 2017; 8: 538–549. https://doi.org/10.3389/fmicb.2017.00538 PMID: 28400769.

43. Elena T, Rosanna DP, Emanuela M, Esposito E, Virginia M, Salvatore C, Anti-inflammatory effects of adrenomedullin on acute lung injury induced by carrageenan in mice. Mediators of Inflamm. 2012; 2012: 717851–717864. https://doi.org/10.1155/2012/717851 PMID: 22685374.

44. Moon SM, Lee SA, Hong JH, Kim JS, Kim DK, Kim CS. Oleamide suppresses inflammatory responses in LPS-induced RAW264.7 murine macrophages and alleviates paw edema in a carrageenan-induced inflammatory rat model. Int Immunopharmacol. 2018; 56: 179–185. https://doi.org/10.1016/j.intimp.2018.01.032 PMID: 29414648.

45. Posadas I, Bucci M, Roviezzo F, Rossi A, Parente L, Sautebin L, et al. Carrageenan-induced mouse paw oedema is biphasic, age-weight dependent and displays differential nitric oxide cyclooxygenase-2 expression. Br J Pharmacol. 2004; 142(2): 331–338. https://doi.org/10.1038/sj.bjp.0705650 PMID: 15155540.

46. Solanki HK, Shah DA, Maheriya PM, Patel CA. Evaluation of anti-inflammatory activity of probiotic on carrageenan-induced paw edema in Wistar rats. Int J Biol Macromol. 2015; 72: 1277–1282. https://doi.org/10.1016/j.ijbiomac.2014.09.059 PMID: 25316426.

47. Ivanovska N, Philippov S. Study on the anti-inflammatory action of Berberis vulgaris root extract, alkaloid fractions and pure alkaloids. Int Immunopharmacol. 1996; 18(10): 553–561. https://doi.org/10.1016/s0192-0561(96)00047-1 PMID: 9080249.

48. Kumar R, Gupta Y, Singh S. Anti-inflammatory and anti-granuloma activity of Berberis aristata DC. in experimental models of inflammation. Indian J Pharmacol. 2016; 48(2): 155–161. https://doi.org/10.4103/0253-7613.178831 PMID: 27114638.

49. Alamgeer NH, Rasool S, Raza SA, Ahmad T, Ahsan H, Mushtaq MN, et al. Anti-inflammatory, analgesic and antipyretic activities of the aqueous methanolic extract of Berberis calliobotrys in albino mice. Acta Pol Pharm. 2016; 73(3): 717–723. PMID: 27505907.