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Chapter

Antioxidant Activity of Nigella sativa Essential Oil

Kehinde Sowunmi and Zeenat Kaka

Abstract

Nigella sativa oils have anti-inflammatory, antibacterial, antifungal, antiparasitic and antiprotozoal, antiviral, cytotoxic, anticancer, neuro-, gastro-, cardio-, and hepatoprotective properties, making them potential treatments for a wide range of illnesses. N. sativa oil also suggests positive benefits on the immunological, respiratory, and reproductive systems in addition to diabetes mellitus (DM), fertility, breast cancer, dyspepsia, osmotic balance, and other conditions. Thymoquinone (TQ) is a suitable target for its potential antibacterial, antimicrobial, anti-inflammatory, chemopreventive, antitumoral, and other actions among the various isolated chemical moieties. The N. sativa oil has been shown in various non-clinical and clinical investigations to benefit health. On the other hand, TQ in several animal experiments is clear to generate no adverse modifications of the body biomarkers; rather, it enhanced health quality. This study presents a more mechanistic review of the constitutions and oil of N. sativa. In conclusion, research on Nigella oil may represent a health breakthrough.

Keywords: Nigella sativa, antioxidant, essential oils, potential treatment, thymoquinone (TQ)

1. Introduction

Nigella sativa L. is a short shrub of the Ranunculaceae botanical family. It is native to Southern Europe, North Africa, and Southeast Asia, and it is grown in a variety of nations across the world. Green leaves and 5–10 petalled rosaceous white, yellow, pink, pastel blue, or purple blooms. The mature fruit bears several small dark black seeds. The oil of N. sativa was widely utilised in traditional Asian and Middle Eastern medicines [1]. However, N. sativa has been used to treat a wide range of ailments affecting the respiratory system, digestive tract, cardiovascular system, kidney, liver, and immunological system. It has long been used to treat weariness and depression. Ailments such as asthma, bronchitis, rheumatism and associated inflammatory illnesses, indigestion, lack of appetite, diarrhoea, dropsy, amenorrhea, dysmenorrhea, worms, and skin eruptions are among the most prevalent traditional applications. It’s both an antiseptic and a local anaesthetic [2].

Protein, fat, carbohydrates, crude fibre, total ash, volatile oil, fatty oil, cellulose, and moisture are all present in black seed oil [3]. The oil is also a good source of minerals including Ca, K, Se, Cu, P, Zn, and Fe, as well as several vitamins like A, B1, B2, and C. Additionally, seeds, roots, and shoots contain both carotenes and vanillic acid.
The primary unsaturated fatty acids include linolic acid, oleic acid, diomolinoleic acid, and eicosadienoic acid, which are found in fatty components. The two primary saturated fatty acids that palmitic acid and stearic acid are a part of our -sitosterol and stigmasterol [2]. According to Gharby et al. [4], other fatty acids include myristic acid, palmitoleic acid, linoleic acid, linolenic acid, arachidonic acid, cholesterol, campes- terol, β-sitosterol, 5-avenasterol, and 7-avenasterol. The alkaloids in the oil are either imidazole ring-bearing alkaloids, pyrazole alkaloids, or isoquinoline alkaloids. Terpenes and saponins are also found in them. Evidence suggests that the most significant active ingredients in *N. sativa* include thymoquinone (TQ ), thymohydroquinone, dithy- moquinone, p-cymene, carvacrol, 4-terpineol, t-anethol, sesquiterpene longifolene, –pinene, and thymol, among others. Carvone, nigellicine, nigellone, citrostradienol,
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cycloeucalenol, gramisterol, lophenol, ostusifoliol, stigmastanol, β-amyrin, butyrospermol, and cycloartenol are the other chemical components [2, 5] bitter principle, tannin, resin, reducing sugars, glycosidal saponin, hederagenin glycoside, esters of unsaturated fatty acids with C15 terpenoids, esters of dehydrostearic and linoleic acid, aliphatic alcohol, –unsaturated hydroxy ketone, 3-O-[12-L-rhamnopyrasyl(12)-D-glucopyranosyl]-L-xylopyranosyl (12)] Stigma-5,22-dien-3-D-glucopyranoside, cycloart-23-methyl-7,20,22-triene-3, 25-diol, nigellidine-4-O-sulphite, 11-methoxy-16, 23-dihydroxy-28-methylolean-12-enoate, N. mines A1, A2, B1, and B2 in addition to A3, A4, and A5 [1, 6–16]. The chemical structures of some important chemical moieties are shown in Figure 1.

2. Activities of N. sativa oil

2.1 Antibacterial agent

According to reports, N. sativa has potent antibacterial activity against both gram-positive and gram-negative microorganisms. It has additive effects with spectinomycin, erythromycin, tobramycin, doxycycline, chloramphenicol, nalidixic acid, ampicillin, lincomycin, and co-trimoxazole and exhibits synergistic effects with streptomycin and gentamycin. It can combat resistant microorganisms, including many gram-positive and gram-negative bacteria that are multi-drug resistant [17]. Manju et al. [18] claim that an oil extract from Nigella can guard against Vibrio parahaemolyticus Dahv2 infection in Artemia spp. TQ has demonstrated anti-methicillin-resistant action in Staphylococcus aureus, according to Hariharan et al. [19].

2.2 Antiviral agent

It has been demonstrated that N. sativa increases the ratio of helper to suppressor T cells in people as well as the activity of natural killer (NK) cells. Otherwise, it is an effective inhibitor of murine CMV and HIV protease. In the latter instance, it was discovered that the generation of interferon-gamma (INF-) led to an increase in the quantity and functionality of M-phi and CD4 + ve T cells [7, 8, 17].

2.3 Antifungal activity

When used against Aspergillus niger, Fusarium solani, and Scopulariopsis brevicaulis, N. sativa’s isolated chemical, TQ, was shown to be more efficient than griseofulvin and amphotericin-B. It also has antifungal action against Candida albicans and Madurella mycetomatis. The TQ also works well against Microsporum spp., Trichophyton spp., and Epidermophyton spp. Additionally, against a variety of clinical isolates, such as dermatophytes, moulds, and yeasts, thymohydroquinone and thymol also displayed an antifungal activity [9, 10, 20]. It is also obvious that black seed oil (10–200 g/mL) acts against Saccharomyces cerevisiae and C. utilise [21].

2.4 Effects on parasites

It has been demonstrated that N. sativa oil has potential against leishmaniasis, miracidia, cercariae, and Schistosoma mansoni. In the latter instance, co-treatment with
praziquantel, a well-known anti-schistosomal and anthelmintic medicine for domestic animals, resulted in a potentiating effect from the oil of the black seed, which demonstrated high efficacy [13–16]. According to Simalango [22], ethanol extract of *N. sativa* (0.5–8%) exhibited anti-Ascaris suum action that was noticeably active.

### 2.5 Effect on wound infection

The ability of *N. sativa* oil to speed up the healing of wounds in mice, farm animals, and human gingival fibroblasts was examined. The accumulation result showed that the absolute difference in WBC counts, local infection and inflammation, bacterial growth and tissue damage, and free radical generation had all decreased. Additionally, transforming growth factor beta and basic fibroblast growth factor levels were found to be higher [17]. Studies using *N. sativa* extracts, seed oil, and TQ have been done on the antioxidant activity of Nigella. The research indicates that oxidative stress may have both potential anti-radical and inhibitory effects. Adenosine deaminase (ADA), catalase (CAT), myeloperoxidase (MPO), lipid peroxidase (LPO), reduced glutathione (GSH), glutathione-S-transferase (GSH-ST), glutathione peroxidase (GPx), superoxide dismutase (SOD), and nitric oxide were among the measures that TQ significantly altered (NO). In addition, it decreased levels of malondialdehyde (MDA), conjugated diene (CGD), tumour necrosis factor-alpha (TNF-α), interferon-gamma (IFN-γ), and prostaglandin (PGE2) rather than interleukin-10 (IL-10) and other pro-inflammatory mediators [2, 23].

### 2.6 Anti-inflammatory diseases

*N. sativa* extracts, seed oil, and TQ may have anti-inflammatory properties, according to research using several animal models. The lowering of NO synthesis, interleukin-1 (IL-1), cyclooxygenase-1 (COX-1), cyclooxygenase-2 (COX-2), histone deacetylase (HDAC), and other pro-inflammatory mediators, including IL-1, IL-6, TNF-, IFN-, and PGE2, is a function of this activity [1, 11]. In mice, topical TQ treatments increased the expression of heme oxygenase-1, NAD(P)H-quione oxidoreductase-1, glutamate cysteine ligase, GSH-ST, and other enzymes, but rat seed oil inhibited COXs and 5-LPO in the pathways of arachidonate metabolism [17]. TQ has also been demonstrated to reduce nuclear translocation and nuclear factor-kappa-B (NF-B) DNA binding in mice by preventing I-phosphorylation B5s and subsequent degradation. TQ also reduced the phosphorylation of p38 mitogen-activated protein kinase, c-Jun-N-terminal kinase (c-JUNK), and protein kinase B (Akt) (MAPK–p38). The inactivation of caspase-1 was followed by the suppression of IL-1 and IL-18 in B16F10 mice whose expression of NLRP3 (NACHT, LRR, and pyrin domain-containing protein 3) was reduced. Additionally, the NLRP3 inflammasome was partially inactivated as a result of TQ's inhibitory action on NF-B and reactive oxygen species (ROS) [17, 23, 24].

### 2.7 Anticancer

The capacity of black seed oil to boost NK cells makes it potentially useful in immune treatment. Oil's constituents, however, may have a carcinogenic impact due to prooxidant effects caused by antioxidants. TQ was also evaluated on a variety of cancer cells generated from mice, indicating its capacity to stop G0/G1 phases of the cell cycle, which coincided with rapid increases in the expression of the cyclin-dependent kinase p16 (CDK-p16) and a drop in cyclin-d1 (dcl-1) protein expression.
in papilloma (SP-1) cell line, and G2/M arrest connected with an increase in the production. The chemopreventive potential of TQ may be attributed to its capacity to reduce cyclin-xl (bcl-xl) protein and enhance the ratio of apoptosis regulator (bcl-4)/cyclin-2 (bax/bcl-2) expression. Additionally, squamous cell carcinoma (SCC-VII), FsaR, and mouse tumour models of fibrosarcoma and SCC were found to exhibit TQ’s anticancer efficacy. TQ significantly increased the sub-G1 population, live/dead cytotoxicity, chromatin condensation, DNA laddering, and Tunel-positive cells in A431 and Hep-2 cells, demonstrating substantial anticancer action through apoptosis. Caspase activation, cell proliferation, cleavage of poly ADP ribose polymerase, and a rise in the bax/bcl-2 ratio were also seen [17]. According to research, TQ caused p53-independent apoptosis in human colon cancer cells, as well as p21 expression, and stopped the S phase of the cell cycle [25]. TQ is a potent down-regulator of NF-B and MMP-9 in Panc-1 cells as well as bcl-2 and an up-regulator of caspase-3 and caspase-9 in gastric cancer cells. It is also an anticancer drug for several cell lines, including MCF-7/Topo breast carcinoma cells. The antitumor action of certain TQ derivatives, including 6-menthoxybutyryl, 6-hencosahexanyl conjugate, 4-acyl hydrazones, and 6-alkyl derivatives, is also visible in cancer cell lines [2].

2.8 Effect on diabetes mellitus

In rats, *N. sativa* was discovered to be crucial in the lowering of blood glucose levels with an increase in insulin and C-peptide levels. TQ lowers tissue MDA levels, DNA damage, mitochondrial vaculization, and fragmentation, and by acting as an antioxidant, it protects the integrity of pancreatic beta-cells. According to a study, TQ clearly raises insulin and Hb levels while also significantly lowering glucose and HbA1c levels. In T2 D rats, *N. sativa* improved bone mass, connection, biomechanical behaviour, and strength in a synergistic manner with parathyroid hormone [26, 27]. Additionally, it is clear that the black seeds are an effective treatment for those with dyslipidemia and the insulin resistance syndrome. Diabetes patient Meriones Shauwi treated with *N. sativa* likewise had an insulin-sensitization effect through increased ACC phosphorylation (primarily MAPK signalling pathway) and muscle GLUT4 content as well as gradual restoration of glycemia [1, 28]. In rats with diabetic mellitus (DM) brought on by streptozotocin, lipid and volatile fractions decreased toxicological and unfavourable effects [29]. Additionally, Heshmati et al. [30] reported that therapy with oil at 3 g three times per day might improve glycemic status and lipid profiles in DM patients (n = 72). When TQ was tested in clonal beta-cells and rodent islets, it had a protective effect with normalisation of chronic malonyl CoA accumulation and elevation of acetyl CoA carboxylase (ACC), fatty acid synthase (FAS), and fatty acid binding proteins (FABPs) following chronic glucose overload, suggesting a modification in beta-cell redox circuitry and enhancing the sensitivity of beta-cell metabolic pathways to glucose and glucose-stimulated insulin secretion (GSIS) under both normal conditions and hyperglycemia [31]. Otherwise, MAPK controls several transcriptional variables, the alteration of which disrupts the cell cycle. Therefore, *N. sativa* and TQ may be effective treatments for both type 1 and type 2 DM patients, as maintaining beta-cell integrity and secreting enough insulin to support glycogenesis and the phosphorylation of elevated blood glucose levels are essential in this context. Otherwise, oxidative stress, illness, and trauma are the other variables that raise blood sugar levels in addition to eaten meals. Therefore, the antioxidant, antibacterial, cytotoxic, and anti-inflammatory properties of *N. sativa* and TQ may be related to each other. Otherwise, lowering HbA1c levels is one of the treatments for retinopathy, nephropathy, and cardiovascular disease.
2.9 Effect on the immune system

*N. sativa* is a demodulator of the production of numerous pro-inflammatory mediators, with an increase in the release of Th2 vs. Th1 cytokines in splenocytes, along with NK antitumor activity. Black seed extracts can regain resistance against granulocyte-dependent *C. albicans*. According to research conducted by the oil, the immunosuppressive cytotoxic impact of typhoid immunisation may result in a decrease in antibody production. Additionally, it is clear that the oxytetracycline (OXT)-induced imbalances in leukocyte, lymphocyte, heterophil: lymphocyte, lysosomal enzyme activity, and reticuloendothelial system function need to be addressed. When pigeons received continuous antibiotic therapy, nevertheless, it had an immuno-protective effect. The black seed oil also has radioprotective properties against the oxidative and immunosuppressive effects of ionising radiation. In addition, *N. sativa* oil administration resulted in a rise in IFN- levels and a considerable reduction in the pathological alterations in the guinea pigs' lungs. Additionally, it works well for allergic diarrhoea [1, 23, 24]. Recent research reveals that seed oil can shield the jejunal mucosa from harm caused by -radiation [26]. After 6 weeks of therapy, Nigella EO in hens at doses between 5 and 20 g/kg (oral feed) enhanced FCR, plasma lipid profile, and antibody-mediated immunity [32]. Additionally, in individuals with Hashimoto's thyroiditis, nigella oil decreased thyroid stimulating hormone (TSH) and anti-thyroid peroxidase antibodies [12, 33].

2.10 Effect on the nervous system (NS)

The methanolic extract of *N. sativa* is an effective analgesic and antidepressant. Additionally, rat brains showed anxiolytic action by elevating serotonin (5-HT) and lowering hydroxy indole acetic acid (5-HIAA) levels [20]. Rats showed improved learning and memory associated with an increase in 5-HT secretion. It may aid in the treatment of anxiety since it increased the levels of tryptophan [20, 23]. In contrast, TQ decreased the generation of NO and MDA while still having a GABA-mediated calming effect in mice [1]. Due to its antioxidant, free radical scavenging, and anti-inflammatory properties, it may have neuroprotective properties.

2.11 Effect on the gastrointestinal tract (GIT) system

TQ is gastroprotective because it increases the quantity and activity of gastric mucin, GSH, total nitric oxide (TNO), and SOD while decreasing stomach acid secretion, acid output (AO), pepsin, mucosal lipid peroxidase (LPO), the proton (H+) pump, MPO, and ulcer index (UI). Prostaglandin (PGD)-mediated and/or via antioxidant and ant secretion pathways were hypothesised to reduce ulcer severity in rats. Rats also showed a decrease in LPO and lactate dehydrogenase (LDH), MPO, MDA, and an increase in GSH, SOD, GPx, and GSH-ST without changing stomach CAT. TQ was discovered to have considerable benefits on inflammatory bowel illnesses, anti-*Helicobacter pylori*, body weight reduction, colitis, and diarrhoea [2].

2.12 Effect on the hepatic system

The hepatoprotective action of *N. sativa* is shown by its effects on the enzymes alanine aminotransferase (ALT), aspartate aminotransferase (AST), LDH, total antioxidant capacity (TAC), CAT, MPO, total oxidative status (TOS), and oxidative
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stress index (OSI). Additionally, GSH and TQ enhanced protein carbonyl content, which reduced protein oxidation and improved the cellular fraction's decreased antioxidant levels [1]. Hepatocytes in mouse TIB-73 cells were shielded against N-acetyl-p-aminophenol (APAP)-induced hepatotoxicity and metabolic abnormalities by N. sativa oil at a concentration of 25–100 g/mL [34]. With an aqueous extract of N. sativa, a comparable activity was also seen by Hamza and Salem Al-Harbi [35]. This activity was assumed to be related to enhancing antioxidant capacity and inhibiting both lipid peroxidation and ROS formation [34]. It is also clear that black seed oil, when administered to rats receiving cisplatin (CP), at a dosage of 2 mg/kg, has hepatoprotective effects via enhancing energy metabolism and fortifying antioxidant defence mechanisms [36].

2.13 Effect on the urinary system

N. sativa with ascorbic acid (Vitamin C) reduced serum creatinine (CK), blood urea nitrogen (BUN), and antioxidant activity in rabbits, resulting in a nephroprotective effect. Otherwise, TQ had an impact on the renal expression of organic ion transporters and proteins linked to multidrug resistance in rats. Rats showed lower expression of OAT1, OAT3, OCT1, and OCT2 and increased protein levels of the efflux transporters MRP2 and MRP4. Along with lowering the tubular necrosis score, N. sativa is effective at lowering levels of CK, urea, MDA, NO, ROS, OSI, and TOS in kidney tissue and blood while increasing TAC, SOD, and GPx. The gentamicin (GM)-induced change in blood CK, BUN, thiobarbituric acid substances (TBARS), and total bilirubin is reversed by TQ. The black seed ethanol extracts showed considerable nephroprotective effect against paracetamol-induced nephrotoxicity in female Wistar Albino rats at doses of 250–100 mg/kg [37]. Otherwise, Erboga et al. [38] have shown that Cd-induced nephroprotective is also detectable in rats.

2.14 Effect on the pulmonary system

Leukotriene-d4 (LT4) is inhibited by both nigellone and TQ in the trachea, where the activity of the former was determined by mucociliary clearance. The peribronchial inflammatory cell infiltration, alveolar septal infiltration, alveolar edema, alveolar exudates, alveolar macrophages, intestinal fibrosis, granuloma, necrosis formation, NOS, and an increase in surfactant protein D in the pulmonary system were all significantly decreased by N. sativa. It is also clear that N. sativa protects the lungs from damage brought on by hypoxia and lung injury. N. sativa puffs have also been shown to have a bronchodilatory impact on PFT values, frequency of asthma symptoms/weakness, chest wheezing, and asthma symptoms [1].

2.15 Effect on the reproductive system

TQ reduced the levels of TAC and MPO in C57BL/6 male mice. Additionally, TQ warned of methotrexate-related occurrences such as intestinal space enlargement, edema, disruption of the somniferous epithelium, and smaller seminiferous tubule diameter. Treatment of 34 infertile men for two months with 2.5 mL black seed oil enhanced their abnormal semen quality without having any negative effects [39]. Black seed oil is a promising therapy for treating male infertility, according to Mahdavi et al. [28] In Sprague-Dawley male and female rats, N. sativa extracts in hexane and methanol significantly reduced fertility. In contrast, N. sativa prevented
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the contraction of uterine smooth muscle in rats and guinea pigs [1, 28]. TQ reduced the number of polycystic ovaries in rats by reducing their exposure to olive oil [40].

2.16 Effect on dyspepsia

A substantial reduction in dyspepsia was seen in individuals (n = 70) with functional dyspepsia who received treatment with 5 mL of Nigella oil (p.o.) daily for 8 weeks [41]. In osmotic balance: Nigella It was determined that black seed oil (22.6 g/25 l) should be used as an alternate therapy to isotonic sodium chloride (0.9% NaCl) solution for the elderly patients (n = 42) after they received treatment for two weeks (Table 1) [43].

<table>
<thead>
<tr>
<th>Chemicals</th>
<th>Dose</th>
<th>Activity</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Essential oil</td>
<td>Antioxidant assays: 5–50 g/L, antimicrobial assays: 0.2–2.0 g/mL</td>
<td>Produced antioxidant activity and shielded Artemia species following experimental <em>Vibrio parahaemolyticus</em> Dahv2 infection.</td>
<td>Manju et al. [42]</td>
</tr>
<tr>
<td>Oil</td>
<td>—</td>
<td>Decreased levels of TG, LDL, and total cholesterol; higher levels of HDL.</td>
<td>Sahebkar et al. [27]</td>
</tr>
<tr>
<td>Oil</td>
<td>400 mg/kg (i.g.) in Wistar albino rats</td>
<td>Reduced glutathione peroxidase (GSH-Px) and superoxide dismutase (SOD) activity were increased and reduced glutathione aldehyde (MDA) levels were decreased in intestinal tissue samples.</td>
<td>Orhon et al. [26]</td>
</tr>
<tr>
<td>Oil</td>
<td>Oil 2 mg/kg (p.o.) in cisplatin (CP) treated mice</td>
<td>Improved energy metabolism and strengthened antioxidant defence mechanisms to produce hepatoprotective effects.</td>
<td>Kehinde et al. [20]</td>
</tr>
<tr>
<td>Oil</td>
<td>Patients with functional dyspepsia 5 ml (p.o.)</td>
<td>Lowered dyspepsia</td>
<td>Mohtashami et al. [41]</td>
</tr>
<tr>
<td>Oil</td>
<td>—</td>
<td>Antioxidant, immunomodulatory, and anti-inflammatory properties; a viable therapy option for male infertility. Potential to combat diabetes mellitus. Interaction with hepat- and kidney protection</td>
<td>Kehinde et al. [20]</td>
</tr>
<tr>
<td>TQ</td>
<td>In β-cells and rodent islets</td>
<td>During chronic glucose overload, a protective activity linked to the stabilisation of chronic malonyl CoA buildup and increase of acetyl CoA carboxylase (ACC), fatty acid synthase (FAS), and fatty acid binding proteins (FABPs). Thus, in both normal circumstances and hyperglycemia, the modified cell redox circuitry and enhanced sensitivity of the metabolic pathways to glucose and glucose-stimulated insulin secretion (GSIS) are present.</td>
<td>Grey et al. [31]</td>
</tr>
</tbody>
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Table 1.
Several new research findings on nigella recipes.
3. Conclusion

One of the potential sources of drugs comes from plants, specifically shrubs. It’s interesting to note that people worldwide are paying a lot of attention to herbal medicines today. Otherwise, traditional medicines continue to rule a certain kingdom of remedies. The excitement for drug researchers comes from the possible and varied activities of a trustworthy source. According to earlier research, *N. sativa* generated notable pharmacological actions, mainly through the use of TQ and its derivatives, nigellone, –hederin, and linoleic acid. Additionally, a few human clinical applications imply that *N. sativa* and its genetic makeup have a safety record. *N. sativa* and its derivatives may be chemically modified to produce useful results for the drug library. It is been found to be safe and healthy in several clinical applications, particularly in anti-fertility studies. Fixed oil of black seed was found to have LD50 values of 26.2–31.6 mg/kg in mice when administered intraperitoneally (i.p.) and orally (p.o.). TQ was found to be more tolerable than the *N. sativa* extract. They may be effective sources of cytoprotective agents due to their substantial antioxidant activity through antiradical, including ROS, direct reduction of oxidizable substrates, and stimulation of cellular antioxidant molecules. Antibiotics or radiation therapy can be used in conjunction with *N. sativa* oil to counteract its cytotoxic, immunosuppressive, and carcinogenic effects. TQ fits within this category, while further research is needed to determine its genotoxic and mutagenic potential.

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