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Investigation of Nitrosamine Impurities in Pharmaceutical Preparations Containing Metformin and Sitagliptin Active Ingredients

Metformin ve Sitagliptin Etkin Maddelerini İçeren Farmasötik Preparatlardaki Nitrozamin Safsızlıklarının Araştırılması

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Özet

Amaç: Bu çalışmada, aktif farmasötik bileşen olarak metformin ve sitagliptin içeren ilaçlardaki N-nitrosodimetilamin (NDMA) ve N-nitrosodietilamin (NDEA) içeriklerinin belirlenmesi amaçlanmıştır.

Yöntem: Metformin ve sitagliptin içeren farmasötik ürünler yerel eczanelerden temin edilmiştir. Bu ilaçlardaki NDMA ve NDEA düzeyleri, headspace gaz kromatografisi-kütle spektrometrisi (HS-GC-MS) kullanılarak analiz edilmiştir.

Bulgular: NDMA konsantrasyonları 0.920 ile 1.571 mg.kg-1 arasında, NDEA konsantrasyonları ise 1.241 ile 2.908 mg.kg-1 arasında saptanmıştır. Her bir tabletteki NDMA miktarları 4.59.10-4 – 1.65.10-3 mg arasında ve NDEA miktarları da 7.94.10-4 – 3.54.10-3 mg arasında tespit edilmiştir.

Sonuç: Elde edilen değerler kabul edilebilir alım limitlerinin (NDMA için 96 ng/gün ve NDEA için 26,5 ng/gün) üzerinde bulunmuştur.

Anahtar Kelimeler: İlaç, headspace GC-MS, N-nitrosodietil amin (NDEA), N-nitrosodimetil amin (NDMA)

Abstract

Aim: In this study, the determination of N-nitrosodimethyl amine (NDMA) and N-nitrosodiethyl amine (NDEA) contents in medicines containing metformin and sitagliptin as active pharmaceutical ingredients was aimed.

Methods: Pharmaceutical products containing metformin and sitagliptin were obtained from local pharmacies. The levels of NDMA and NDEA in these medicines were analyzed using headspace gas chromatography–mass spectrometry (HS-GC-MS).

Results: NDMA concentrations were found to range between 0.920 and 1.571 mg.kg-1, while NDEA levels ranged from 1.241 to 2.908 mg.kg-1. On a per-tablet basis, the detected NDMA amounts ranged from 4.59.10-4 to 1.65.10-3 mg, and NDEA amounts ranged from 7.94.10-4 to 3.54.10-3 mg.

Conclusion: The values obtained were found to be above the acceptable intake limits (96 ng/day for NDMA and 26.5 ng/day for NDEA).

Key Words: Drug, headspace GC-MS, N-nitrosodiethyl amine (NDEA), N-nitrosodimethyl amine (NDMA)

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1.Introduction

Nitrosamines are a class of organic compounds with a general chemical structure expressed as $R_1R_2N–N=O$ consisting of a nitroso group attached to a dialkylamine. In this structure, R_1 and R_2 represent alkyl or aryl groups (1). N-nitrosamines are formed when an amine reacts with a nitrosating agent. The activity of nitrosating agents is important in the formation of nitrosamines. Furthermore, the structure of amines, the concentrations of reactants, and the presence of inorganic ions also affect nitrosamine formation. Nitrosamines can also form in different environments, depending on reaction conditions and the factors that affect these reactions. Nitrosamines are found in food products such as salami, sausage, meat, fish, chicken, beer, margarine, vegetable oils, and cheese; in the human body, in animal tissues and organs; in biological samples such as saliva, blood, urine, and serum; in environmental samples such as water, air, soil, and cigarette smoke; and in industrial chemicals such as pharmaceuticals, pesticides, and herbicides (2).

The link between cancer and nitrosamines was first established in the 1950s. Numerous tumor formations were observed in mink fed nitrite-containing feed. This event led to an understanding of the relationship between tumor formation and nitrosamines (3). Studies conducted on 40 different types of experimental animals have shown that nitrosamines are carcinogenic. Furthermore, more than 300 different types of nitrosamines have been shown to cause carcinogenic effects (4). Following all these studies, the International Agency for Cancer Research (IARC) classified nitrosamines as carcinogenic to humans in 1987 (5).

Metformin is considered the cornerstone therapy for type 2 diabetes mellitus and is extensively prescribed worldwide. Metformin is the most widely used drug worldwide for the treatment of type 2 diabetes (6). Dimethylamine hydrochloride is defined as an impurity in metformin and is generally found in metformin in the range of 15 to 230 ppm, with a limit of 500 ppm. Some excipients used in metformin formulations contain approximately 1–2 ppm nitrite; therefore, there is a potential for the formation of N-nitrosodimethylamine (NDMA) (7).

Sitagliptin is a potent and selective dipeptidyl peptidase-4 inhibitor approved by the EMA and the Food and Drug Administration (FDA) for use in the treatment of adult patients with type 2 diabetes (8). Sitagliptin contains dimethylformamide, diisopropylethylamine, and 4-dimethylaminopyridine, which are precursors of some types of nitrosamines. These nitroso impurities can occur as process impurities during the manufacturing process of pharmaceutical materials (9).

Nitrosamines in pharmaceuticals are caused by contaminated starting materials and intermediates, reagents, solvents, catalysts, errors in the production process, recovered and recycled materials, and manufacturers lacking sufficient process knowledge. Furthermore, nitrosamines can also form in pharmaceuticals after production, depending on storage conditions, packaging materials, and the degradation processes of the active ingredients. Nitrogen oxides can form when nitrocellulose-containing pharmaceutical packaging is sealed by heat. This also creates a breeding ground for nitrosamine formation (10).

The worldwide awareness regarding nitrosamine contamination in medicinal products markedly increased following the identification of NDMA in certain antihypertensive drug batches in 2018. These assessments revealed that nitrosamine impurities were not limited to valsartan but were also present at unacceptable levels in other widely used medicines, including ranitidine, angiotensin II receptor blockers, metformin, and nizatidine. Furthermore, NDMA levels were shown to increase during storage at room temperature, and water used in active pharmaceutical ingredient (API) manufacturing may contain nitrite or nitrosamines due to environmental contamination. In response, the EMA's Committee for Medicinal Products for Human Use issued an opinion in 2020 emphasizing the need to implement risk assessment and control strategies to minimize nitrosamine formation arising from raw materials, APIs, or manufacturing processes, in line with ICH M7 (R1) on mutagenic impurities. To limit potential carcinogenic risk to humans, N-nitrosamines with TD50 values below 1.5 mg/kg/day are classified as compounds of concern, and acceptable intake limits have been established for each specific nitrosamine (11-13).

In recent years, the detection of nitrosamine impurities in pharmaceutical products used in many countries has become a serious concern due to the potential risks these compounds pose to human health. These developments have led researchers to focus on the mechanisms of nitrosamine formation, exposure levels, and risk management strategies; particularly abroad, an increasing number of scientific studies and regulatory frameworks have been established in this area (14-25). However, the lack of systematic, comprehensive scientific studies on the presence and levels of nitrosamine impurities in pharmaceutical products in our country until recently indicates a significant knowledge gap in this field. In our previous study, the levels of N-nitrozodimethylamine and N-nitrozodiethylamine (NDEA) in cardiovascular drugs containing sartan, commonly prescribed in our country, were analyzed using the headspace GC-MS method; the results revealed that the nitrosamine levels per tablet exceeded the acceptable daily intake limits set by international authorities. These findings suggest that nitrosamine impurities may not be limited to sartan group drugs and may also be present in other pharmaceutical products commonly used to treat chronic diseases. Accordingly, the aim of the present study was to determine the levels of NDMA and NDEA in metformin- and sitagliptin-containing drugs, which are commonly used to treat type 2 diabetes.

2. Method

Nitrosamine analyses were performed using a Headspace Gas Chromatography–Mass Spectrometry (HS-GC-MS) system. An Agilent 7697A Headspace sampler was used for sample preparation and introduction. Samples were incubated at an oven temperature of 120 °C, while the sample line and transfer line temperatures were maintained at 125 °C and 130 °C, respectively. The system pressure was set at 103 kPa, and an equilibration time of 15 min was applied to ensure adequate partitioning of volatile analytes into the headspace phase. Gas chromatographic separation was carried out using an Agilent 7890A gas chromatograph equipped with a DB-WAX capillary column (50 m × 0.20 mm i.d., 0.20 µm film thickness). Helium was used as the carrier gas at a constant flow rate of 2 mL.min-1. The injector and detector temperatures were maintained at 240 °C. The oven temperature program was initiated at 40 °C and held for 2 min, followed by a ramp to 120 °C at 10 °C min-1, with a 2 min hold. Subsequently, the temperature was increased to 230 °C at a rate of 25 °C.min-1 and maintained for 5 min. Mass spectrometric detection was performed using an Agilent 5975C mass selective detector operating in electron impact (EI) ionization mode at 70 eV. Data acquisition was performed in selected-ion monitoring (SIM) mode to achieve high sensitivity and selectivity for nitrosamine determination. For N-nitrosodimethylamine (NDMA), the ions monitored were m/z 74, 42, and 43, whereas for N-nitrosodiethylamine (NDEA), the ions monitored were m/z 102, 57, 56, 44, and 42. Under these optimized analytical conditions, reliable and sensitive determination of volatile nitrosamines was achieved.

2.1. Research Questions / Hypothesis

The primary research question of this study is whether nitrosamine impurities (NDMA and NDEA) can be detected in pharmaceutical products available on the Turkish market and whether the developed HS-GC–MS method enables reliable determination of these compounds at trace levels. It is hypothesized that the optimized headspace GC–MS method provides sufficient sensitivity and selectivity for the accurate and reproducible quantification of NDMA and NDEA at low concentrations.

2.2. Population and Sample

The study population consists of pharmaceutical products available in the Turkish market. The sample set includes selected pharmaceutical products obtained from different retail pharmacies. The samples were randomly collected and represent different manufacturers and batch numbers to ensure variability.

2.3. Data Collection and Analytical Tools

Data were obtained using a headspace GC–MS system. A total of 28 commercially available pharmaceutical products containing metformin and sitagliptin as active pharmaceutical ingredients (APIs) were obtained from local pharmacies and analyzed in this study. For sample preparation, 0.1 g of each pharmaceutical sample was dissolved in 2 mL of dimethyl sulfoxide (DMSO). The resulting mixture was vortexed at room temperature until a homogeneous solution was obtained. Subsequently,

the prepared solutions were transferred into headspace vials for analysis. The analyses were performed using an Agilent gas chromatography system coupled with a mass spectrometer. Mass spectrometric detection was conducted in selected ion monitoring (SIM) mode, and characteristic ions for NDMA and NDEA were used for quantitative evaluation. A series of standard solutions for N-nitrosodimethylamine and N-nitrosodiethylamine were prepared in methanol over the concentration range of 0.05 – 20 $mg.L^{-1}$. These standards were used to establish calibration curves for quantitative analysis. Calibration plots were generated by correlating the peak areas obtained from HS-GC-MS analysis with the analyte concentrations.

2.4. Ethical Considerations

This study does not involve human or animal subjects. The analyzed pharmaceutical products were commercially available, and no personal or clinical data were used. Therefore, ethical approval was not required.

2.5. Limitations of the Study

The main limitations of this study include the limited number of analyzed samples and the focus on only two nitrosamine compounds (NDMA and NDEA). Additionally, variations related to different production batches and long-term storage conditions were not extensively investigated.

2.6. Data Analysis and Evaluation

Chromatographic data were processed using the instrument software. Quantitative results were obtained based on peak area measurements and corresponding calibration curves. Method performance was evaluated for linearity (R^2), limit of detection (LOD), limit of quantification (LOQ), and reproducibility. Excellent linearity was obtained for both analytes, with correlation coefficients of 0.999 for NDMA and NDEA. The LOD and LOQ were determined to be 0.004 mg.L-1 and 0.013 mg.L-1 for NDMA, respectively, while the corresponding values for NDEA were 0.001 mg. L-1 and 0.003 mg.L-1. All analyses were performed in triplicate, and the results were reported as mean values.

3. Results

The concentrations of N-nitrosodimethylamine and N-nitrosodiethylamine detected in the analyzed pharmaceutical samples are presented in Table 1.

Table 1. Amounts of NDMA and NDEA detected in drug samples (N=3)

Active Ingredient of the Drug	Amount of the Active Ingredient in the Drug (mg)	NDMA (mg kg^{-1})	NDEA (mg kg^{-1})
Sitagliptin	100	1.147 ± 0.01	2.107 ± 0.01
Sitagliptin	100	1.103 ± 0.01	2.529 ± 0.01
Metformin	850	1.076 ± 0.01	2.041 ± 0.01
Metformin	1000	0.947 ± 0.01	2.266 ± 0.01
Metformin	500	1.571 ± 0.01	2.916 ± 0.01
Metformin	1000	1.179 ± 0.01	2.738 ± 0.01
Metformin	850	1.111 ± 0.01	1.955 ± 0.01
Metformin	1000	1.476 ± 0.01	2.755 ± 0.01
Metformin	850	1.226 ± 0.01	2.579 ± 0.01
Metformin	1000	1.230 ± 0.01	3.093 ± 0.01
Metformin	1000	0.995 ± 0.01	2.555 ± 0.01
Metformin	850	1.196 ± 0.01	2.140 ± 0.01
Metformin	1000	1.347 ± 0.01	2.908 ± 0.01
Metformin	500	0.920 ± 0.01	1.241 ± 0.01
Metformin	850	1.039 ± 0.01	2.094 ± 0.01
Metformin	1000	1.073 ± 0.01	2.285 ± 0.01
Metformin	850	0.972 ± 0.01	2.357 ± 0.01
Metformin	1000	1.241 ± 0.01	2.715 ± 0.01
Metformin	850	1.178 ± 0.01	2.551 ± 0.01
Metformin	1000	1.252 ± 0.01	1.962 ± 0.01
Metformin	850	1.353 ± 0.01	2.329 ± 0.01
Metformin	1000	0.952 ± 0.01	2.309 ± 0.01
S + M	50/500	1.039 ± 0.01	2.380 ± 0.01
S + M	50/850	1.506 ± 0.01	2.587 ± 0.01
S + M	50/1000	1.012 ± 0.01	2.159 ± 0.01
S + M	50/500	1.489 ± 0.01	2.040 ± 0.01
S + M	50/850	1.239 ± 0.01	2.877 ± 0.01
S + M	50/1000	1.517 ± 0.01	2.343 ± 0.01

S: Sitagliptin M: Metformin

In sitagliptin-containing products (100 mg), mean NDMA concentrations ranged from 1.103 to 1.147 mg.kg-¹, while NDEA concentrations ranged from 2.107 to 2.529 mg.kg-¹. In metformin-containing formulations, NDMA concentrations were found between 0.920 and 1.571 mg.kg-¹ (500 mg), 0.972 and 1.353 mg.kg-¹ (850 mg), and 0.947 and 1.476 mg.kg-¹ (1000 mg). Corresponding NDEA concentrations ranged from 1.241 to 2.916 mg.kg-¹, 1.955 to 2.579 mg.kg-¹, and 1.962 to 3.093 mg.kg-¹, respectively. For combination products containing sitagliptin and metformin (S/M), NDMA concentrations ranged from 1.039 to 1.517 mg.kg-¹, while NDEA concentrations ranged from 2.040 to 2.877 mg.kg-¹ depending on formulation strength.

The amounts of NDMA and NDEA per tablet are presented in Table 2. NDMA levels ranged from $4.59.10^{-4}$ to $2.07.10^{-3}$ mg per tablet, whereas NDEA levels ranged from $8.62.10^{-4}$ to $3.54.10^{-3}$ mg per tablet across all formulations. All calculated per-tablet values exceeded the acceptable daily intake (ADI) limits of 96 ng day-¹ for NDMA and 26.5 ng day-¹ for NDEA.

Table 2. Amounts of NDMA and NDEA detected in each tablet of the drug samples

Active Ingredient of the Drug	Amount of the Active Ingredient in the Drug (mg)	NDMA (mg/tablet)	NDEA (mg/tablet)
Sitagliptin	100	$4.69.10^{-4}$	$8.62.10^{-4}$
Sitagliptin	100	$4.59.10^{-4}$	$1.05.10^{-3}$
Metformin	850	$9.85.10^{-4}$	$1.87.10^{-3}$
Metformin	1000	$1.00.10^{-3}$	$2.41.10^{-3}$
Metformin	500	$1.65.10^{-3}$	$3.06.10^{-3}$
Metformin	1000	$1.29.10^{-3}$	$2.99.10^{-3}$
Metformin	850	$9.98.10^{-4}$	$1.76.10^{-3}$
Metformin	1000	$1.58.10^{-3}$	$2.95.10^{-3}$
Metformin	850	$1.22.10^{-3}$	$2.57.10^{-3}$
Metformin	1000	$1.41.10^{-3}$	$3.54.10^{-3}$
Metformin	1000	$1.19.10^{-3}$	$3.05.10^{-3}$
Metformin	850	$1.18.10^{-3}$	$2.11.10^{-3}$
Metformin	1000	$1.56.10^{-3}$	$3.37.10^{-3}$
Metformin	500	$5.88.10^{-4}$	$7.94.10^{-4}$
Metformin	850	$1.13.10^{-3}$	$2.29.10^{-3}$
Metformin	1000	$1.36.10^{-3}$	$2.90.10^{-3}$
Metformin	850	$1.06.10^{-3}$	$2.57.10^{-3}$
Metformin	1000	$1.52.10^{-3}$	$3.34.10^{-3}$
Metformin	850	$1.21.10^{-3}$	$2.62.10^{-3}$
Metformin	1000	$1.52.10^{-3}$	$2.39.10^{-3}$
Metformin	850	$1.40.10^{-3}$	$2.41.10^{-3}$
Metformin	1000	$1.13.10^{-3}$	$2.74.10^{-3}$
S + M	50/500	$7.82.10^{-4}$	$1.79.10^{-3}$
S + M	50/850	$1.73.10^{-3}$	$2.97.10^{-3}$
S + M	50/1000	$1.37.10^{-3}$	$2.91.10^{-3}$
S + M	50/500	$1.07.10^{-3}$	$1.47.10^{-3}$
S + M	50/850	$1.42.10^{-3}$	$3.31.10^{-3}$
S + M	50/1000	$2.07.10^{-3}$	$3.20.10^{-3}$

tagliptin M: Metformin (Values were calculated using the mean concentrations obtain from triplicate analyses of each sample presented in Table 1.)

To estimate potential patient exposure, a daily intake scenario assuming the consumption of three tablets per day was applied to each pharmaceutical product. Exposure calculations were performed based on a reference body weight of 70 kg. Using the equations presented below [28], both maximum exposure levels and corresponding hazard index values were determined. The calculated data are summarized in Tables 3 and 4. The acceptable daily intake was set at 9.6 × 10^-5 mg/day for NDMA and 2.6 × 10^-5 mg/day for NDEA.

$$\text{Maximum exposure rate} = \frac{\text{Amount of nitrosamine in the drug} \times \text{weight of the drug} \times \text{daily dose}}{\text{Body weight}}$$

$$Hazard\ index = \frac{Maximum\ exposure\ rate}{TDI}$$

Table 3. The maximum exposure rates

Active Ingredient of the Drug	Amount of the Active Ingredient in the Drug (mg)	NDMA (mg/kg/day)	NDEA (mg/kg/day)	Total Nitrozamine (mg/kg/day)
Sitagliptin	100	$5.48.10^{-6}$	$1.01.10^{-5}$	$1.56.10^{-5}$
Sitagliptin	100	$5.46.10^{-6}$	$1.25.10^{-5}$	$1.80.10^{-5}$
Metformin	850	$2.58.10^{-5}$	$4.89.10^{-5}$	$7.47.10^{-5}$
Metformin	1000	$3.04.10^{-5}$	$7.34.10^{-5}$	$1.04.10^{-4}$
Metformin	500	$4.94.10^{-5}$	$9.17.10^{-5}$	$1.41.10^{-4}$
Metformin	1000	$4.03.10^{-5}$	$9.35.10^{-5}$	$1.34.10^{-4}$
Metformin	850	$2.56.10^{-5}$	$4.52.10^{-5}$	$7.08.10^{-5}$
Metformin	1000	$4.84.10^{-5}$	$9.04.10^{-5}$	$1.39.10^{-4}$
Metformin	850	$3.48.10^{-5}$	$7.33.10^{-5}$	$1.01.10^{-5}$
Metformin	1000	$4.61.10^{-5}$	$1.16.10^{-4}$	$1.62.10^{-4}$
Metformin	1000	$4.06.10^{-5}$	$1.04.10^{-4}$	$1.45.10^{-4}$
Metformin	850	$3.33.10^{-5}$	$5.95.10^{-5}$	$9.28.10^{-5}$
Metformin	1000	$5.17.10^{-5}$	$1.12.10^{-4}$	$1.64.10^{-4}$
Metformin	500	$1.07.10^{-5}$	$1.45.10^{-5}$	$2.52.10^{-5}$
Metformin	850	$3.53.10^{-5}$	$7.15.10^{-5}$	$1.07.10^{-4}$
Metformin	1000	$4.93.10^{-5}$	$1.05.10^{-4}$	$1.54.10^{-4}$
Metformin	850	$3.30.10^{-5}$	$8.01.10^{-5}$	$1.13.10^{-4}$
Metformin	1000	$5.34.10^{-5}$	$1.17.10^{-4}$	$1.70.10^{-4}$
Metformin	850	$3.55.10^{-5}$	$7.68.10^{-5}$	$1.12.10^{-4}$
Metformin	1000	$5.28.10^{-5}$	$8.31.10^{-5}$	$1.36.10^{-4}$
Metformin	850	$4.14.10^{-5}$	$7.13.10^{-5}$	$1.13.10^{-4}$
Metformin	1000	$3.83.10^{-5}$	$9.28.10^{-5}$	$1.31.10^{-4}$
S + M	50/500	$1.68.10^{-5}$	$3.85.10^{-5}$	$5.53.10^{-5}$
S + M	50/850	$5.67.10^{-5}$	$9.74.10^{-5}$	$1.54.10^{-4}$
S + M	50/1000	$5.28.10^{-5}$	$1.12.10^{-4}$	$1.65.10^{-4}$
S + M	50/500	$2.20.10^{-5}$	$3.02.10^{-5}$	$5.22.10^{-5}$
S + M	50/850	$4.67.10^{-5}$	$1.09.10^{-4}$	$1.56.10^{-4}$
S + M	50/1000	$8.08.10^{-5}$	$1.25.10^{-4}$	$2.06.10^{-4}$

S: Sitagliptin M: Metformin (Values were calculated using the data presented in Table 2.)

Table 4. Hazard indexes

Active Ingredient of the Drug	Amount of the Active Ingredient in the Drug (mg)	NDMA	NDEA
Sitagliptin	100	0.057	0.381
Sitagliptin	100	0.057	0.472
Metformin	850	0.269	**1.845**
Metformin	1000	0.317	**2.770**
Metformin	500	0.514	**3.460**
Metformin	1000	0.420	**3.528**
Metformin	850	0.267	**1.706**
Metformin	1000	0.504	**3.411**
Metformin	850	0.362	**2.766**
Metformin	1000	0.480	**4.377**
Metformin	1000	0.423	**3.924**
Metformin	850	0.347	**2.245**
Metformin	1000	0.538	**4.226**
Metformin	500	0.111	0.547
Metformin	850	0.368	**2.698**
Metformin	1000	0.513	**3.962**
Metformin	850	0.344	**3.023**
Metformin	1000	0.556	**4.415**
Metformin	850	0.370	**2.898**
Metformin	1000	0.550	**3.136**
Metformin	850	0.431	**2.690**
Metformin	1000	0.399	**3.502**
S + M	50/500	0.175	**1.453**
S + M	50/850	0.591	**3.675**
S + M	50/1000	0.550	**4.226**
S + M	50/500	0.229	**1.140**
S + M	50/850	0.486	**4.113**
S + M	50/1000	0.842	**4.717**

S: Sitagliptin M: Metformin (Values were calculated using the data presented in Table 3.)

4. Discussion

The results clearly indicate the presence of both NDMA and NDEA in all analyzed pharmaceutical samples, with NDEA consistently detected at higher concentrations than NDMA. This trend suggests that NDEA may represent a more significant contamination concern in the investigated products.

Although NDMA has been widely studied and regulated due to its carcinogenic potential, the elevated levels of NDEA observed in this study highlight the need for increased attention toward this compound. The fact that most NDEA-related hazard index values exceed the safety threshold (HI > 1) indicates a potential health risk under the defined exposure conditions.

In contrast, NDMA-related hazard index values remained below 1 for all samples, suggesting that, within the assumptions of this study, NDMA exposure alone may not pose an immediate risk. However, considering the cumulative exposure to multiple nitrosamines, the combined effect should not be overlooked.

The detection of nitrosamines in both single-ingredient and combination drug products suggests that contamination may arise from multiple sources, including raw materials, manufacturing processes, or storage conditions. This is consistent with previous reports in the literature indicating widespread nitrosamine contamination across various pharmaceutical classes.

Given that all measured values exceeded the acceptable daily intake limits, even at the single-tablet level, these findings underscore the need for stricter regulatory oversight and quality control measures.

5. Conclusion

In this study, NDMA and NDEA impurities were quantitatively determined in selected pharmaceutical products commonly used in Türkiye. The findings demonstrate that nitrosamine contamination is present across different formulations and dosage strengths. The results indicate that NDEA poses a greater potential health risk than NDMA based on hazard index evaluation. These findings underline the necessity for comprehensive monitoring strategies and further investigation into the sources of nitrosamine formation. This study provides both a scientific reference and a methodological framework for future research on the detection and risk assessment of nitrosamine impurities in pharmaceutical products.

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