

# Can Pistachio Nuts Replace Melatonin Tablets?

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## Abstract

**Background:** Melatonin is a component of many plants for example pineapples, grapes or nuts. It is proven that melatonin can be found in pistachios, however, data on its quantitative content varies widely. We determined melatonin content in pistachio nuts (*pistacia vera*) widely available in stores in Poland.

**Methods:** We also examined the concentration of melatonin in the saliva of volunteers (n=6) after consuming 25 grams of peeled pistachios. For the analysis, we used liquid chromatography linked to mass spectrometry (LC-MS/MS).

**Results:** Mean melatonin content in pistachios was  $69.4 \pm 3.8$  ng/g which corresponds to  $1.7 \pm 0.1$  µg/25 g. The concentration of melatonin in saliva after eating 25 g of pistachio was not significantly different from the baseline concentration.

**Conclusions:** Melatonin contained in pistachios can therefore not exert any biological effect and this means that pistachios cannot replace melatonin tablets.

**Keywords:** mass spectrometry; melatonin concentration; pistachio nuts; saliva

## 1.Introduction

There are many reports of the presence of melatonin in various plants, including fruits, nuts and other plant-based foods. Small amounts of melatonin have been determined in pineapples, apples, strawberries, bananas, tomatoes, grapes, among others, with contents ranging from 0.14-14.8 ng/g [1]. The melatonin content of various nuts is 0.1-3.3 ng/g [2]. The highest melatonin content of 230 µg/g (230,000 ng/g) was described in Iranian pistachios [3]. This is at least several thousand times higher than in other plants, and much higher than reported in other studies on pistachios (0.1-12.0 ng/g) [2,4]. A melatonin content of 230 µg/g would mean that after consuming just 10 g of pistachios, one would get a therapeutic dose of melatonin (2.3 mg).

## 2. Results

Determination of melatonin content in pistachios was carried out 3 times from the same pistachio sample. The average melatonin content in pistachios was  $69.4 \pm 3.8$  ng/g which corresponds to  $1.7 \pm 0.1$  µg/25 g. This amount is about 500-1000 times lower than that found in medicinal products or dietary supplements containing melatonin and should not affect the concentration of melatonin in saliva. The Table 1 below shows the results of salivary melatonin concentrations after consuming about 25 grams of peeled pistachios. The study was conducted on a group of n=6 volunteers, the samples were collected between 11 a.m. - 3 p.m.

Time after pistachios consumption [h]	Melatonin concentration in saliva [pg/mL]
0	$1.2 \pm 2.9$
0.5	$2.2 \pm 5.4$
1	$0.0 \pm 0.0$
2	$1.2 \pm 2.9$
3	$1.7 \pm 4.1$
4	$0.0 \pm 0.0$

Table 1: Mean melatonin concentration (n=6) after pistachios intake.

## 3. Discussion

The results of the analysis indicate that the concentration of melatonin in saliva after consuming 25 g of pistachios is practically unchanged, thus no any biological effect of melatonin is expected after consuming pistachios. The bioavailability of melatonin after oral administration varies between 2.5 and 33% [2], due to a strong first-pass effect (~85%) and limited absorption [2,5]. The amount of melatonin produced by humans is about 40 µg, although this is individually variable (10–80 µg) [2,6,7]. Thus consumption of a portion of pistachios containing about 2 µg of melatonin may not induce any clinically significant biological effects.

#### 4. Materials and Methods

The purpose of the study was to examine the melatonin content of real pistachios (*pistacia vera*) commonly available for sale in Poland. It was also checked how the concentration of melatonin in saliva changes in volunteers who consumed 25 g of pistachios. Determination of melatonin concentration was carried out using a previously developed and validated method in our laboratory using liquid chromatography coupled to tandem mass spectrometry (LC-MS/MS, Shimadzu, Kyoto, Japan) (see Dermanowski et al. 2022) [8]. We used pistachios in shells purchased in a common retail store in Poland. To determine the melatonin content, pistachios were peeled and crushed using a ceramic mortar. 1 gram of crushed pistachios was placed in each glass tube and poured in 5 mL of water (LC-MS grade, VWR International). The samples were sonicated in a water bath for 60 minutes. The next step was to collect the water extract from glass tubes and clean it using 13 mm PES, 0.22 µm syringe filters (LABFIL, ALWSCI Group, China). The purified extract was placed in 2 mL tubes and centrifuged for 10 minutes at 10,000 x g to obtain a clear supernatant without large solid particles. 300 µL were taken from the centrifuged samples for further processing: an internal standard (melatonin-D4, Cayman Chemicals) was added, extracted for 20 minutes with dichloromethane (HPLC grade, VWR International), and evaporated. The extract was dissolved in 5 mM ammonium acetate and analyzed (details in Dermanowski et al. 2022) [8]. The determination of melatonin concentration in saliva was carried out on volunteers. Each volunteer (n=6) received 50 g of un-peeled pistachios to eat, which corresponded to approximately 25 g of peeled nuts. Saliva was collected using cotton Salivettes® (Sarstedt, Nümbrecht, Germany), at time 0 (before eating pistachios) and after 0.5, 1, 2, 3, 4 hour. Results below LLOQ (5 pg/mL) were treated as 0. Saliva was tested according to the method described in our previous work (details in Dermanowski et al. 2022) [8].

#### 5. Conclusions

In conclusion, real pistachios (*pistacia vera*) contain a small amount of melatonin and their consumption does not significantly affect the concentration of melatonin in saliva.

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#### Abbreviations

The following abbreviations are used in this manuscript:

LLOQ

LC-MS

HPLC Lower Limit of Quantification

Liquid Chromatography – Mass Spectrometry

High Performance Liquid Chromatography

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