ACCEPTED VERSION

David J Kennaway
Are the proposed benefits of melatonin-rich foods too hard to swallow?
Critical Reviews in Food Science and Nutrition, 2017; 57(5):958-962

"This is an Accepted Manuscript of an article published by Taylor & Francis in Critical Reviews in Food Science and Nutrition on 15 May 2015 available online: http://dx.doi.org/10.1080/10408398.2014.962686

Are the proposed benefits of melatonin-rich foods too hard to swallow?

David J Kennaway[1,*]

[1]Robinson Research Institute, School of Paediatrics and Reproductive Health, University of Adelaide, Adelaide, South Australia, Australia, 5005

[*]Corresponding Author Email: david.kennaway@adelaide.edu.au

Keywords

Pineal gland hormone; Functional food; Bioavailability; Pharmacokinetics; Antioxidant

Abstract

Melatonin has been proposed as a potent anti-oxidant and its presence in many plants and foods has been suggested to be beneficial for health. Indeed, the concentrations of melatonin in blood, and the melatonin metabolite 6 sulphatoxymelatonin in urine, have been found to increase significantly after ingestion of melatonin rich foods. . In this review the studies have been critically evaluated in light of the reported plant melatonin concentrations and our knowledge of pharmacokinetics of orally administered pure melatonin. In the case of studies involving measurement of plasma melatonin following ingestion of beer or fruits the reported increases in melatonin are not consistent with the amount of melatonin ingested. Similarly the amount of melatonin metabolite excreted following ingestion of melatonin rich foods greatly exceed the amount ingested. It is concluded that studies reporting the appearance of melatonin in blood and its metabolites in urine following ingestion of melatonin rich foods are flawed. While there may be health benefits for certain foods it is difficult to accept that they are due to their low melatonin content.

Melatonin (N-acetyl 5-methoxy tryptamine) is an indole hormone produced in the pineal gland of vertebrates from the amino acid tryptophan. In animals it appears to have many functions, but the main ones revolve around the control of reproductive function in many species (Boden, et al., 2013), including its seasonality (Dardente, 2012) and facilitating the onset of sleep in humans (Keijzer, et al., 2014). A key feature of its production is that it is primarily produced and secreted during darkness, leading to it being called the "hormone of darkness". During the mid 1990s in the USA, melatonin appeared on pharmacy and supermarket shelves as a dietary supplement and there was enormous interest in and promotion of its effects as a new drug that could apparently prevent aging, cure cancer, improve your sex life, etc. (Pierpaoli & Regelson, 1995). It is still available as a supplement in 0.5 - 5 mg doses in the USA, but its sale has always been restricted in other jurisdictions. When used as a dietary supplement or as a drug, it is generally taken to advance the time of sleep or assist in the recovery from jet lag.

In recent years it has emerged that melatonin is also synthesised in a wide range of plants where physiologically it may also have a role as a hormone or as an anti-oxidant (Tan, et al., 2012). In animals, melatonin acts on membrane bound G-protein-coupled receptors, designated MT1 and MT2 receptors and coded by the MTRN1A and MTRN1B genes (Dubocovich, et al., 2010). These are very high affinity receptors with dissociation constants in the low picomolar range.

Endogenous melatonin is metabolised by CYP1A2, sulpho- and glucuronyl-transferase enzymes (Ma, et al., 2005) in the liver to 6-hydroxymelatonin and 6-sulphatoxy melatonin (aMT.6S) or 6-glucuronyl melatonin and then excreted. It has been estimated that 90-95% of administered

melatonin is excreted as these metabolites (Jones, et al., 1969). An additional potential metabolic pathway was proposed to involve oxidation by indoleamine 2,3-dioxygenase giving rise to N1-acetyl-N2-formyl-5-methoxykynurenamine, which is then de-formylated to N1-acetyl-5-methoxykynurenamine (Hirata, et al., 1974). *In vitro*, this transformation can be achieved by other enzymes or chemically. Interestingly these metabolites have not been detected in human urine following large oral doses (Li, et al., 2013). Nevertheless, melatonin has been proposed to be a potent anti-oxidant, protecting cells from reactive oxygen species damage and there have been an extraordinary number of studies published on the potential health benefits of this role of melatonin, particularly when administered in milligram doses (Reiter, et al., 2010). In this review I critically evaluate the evidence that ingestion of melatonin rich foods results in physiologically relevant amounts of melatonin being absorbed and distributed throughout the body of humans.

Melatonin in foods

Since the first report of the detection of melatonin in food (Hattori, et al., 1995), it has been measured in a very wide range of plants and plant products. In Table 1 the contents of melatonin in selected foods are listed; for more extensive lists see (Huang & Mazza, 2011; Paredes, et al., 2009). It is clear that measuring melatonin in plants and foods presents important methodological issues with respect to recovery and detection of low levels of melatonin. In Table 1 it is clear that even for the same type of food, the reported content can vary more than 10 fold between studies. In the case of cherry concentrate, one study did not detect melatonin (Kocadagli, et al., 2014) while another reported 1,420,000 pg/g (Howatson, et al., 2012). (Note that all melatonin concentrations used in this review are shown as pg/g or pg/ml to facilitate easy comparisons between studies).

Oral administration of pure melatonin

Melatonin has been investigated over many years for its effects on sleep and temperature regulation and as a consequence there have been several pharmacokinetic studies of the effects of oral administration on plasma concentrations. As summarised recently, the half-life of oral administered melatonin is less than 60 minutes and the volume of distribution approximately 1000L (Peng, et al., 2013). The bioavailability of an oral fast-release preparation has been reported to be approximately 15% (DeMuro, et al., 2000; Fourtillan, et al., 2000), with men having poorer bioavailability (8.6 ± 3.9%) than women (16.8 ± 12.7%) (Fourtillan, et al., 2000). Using this information we can estimate the expected plasma concentration of melatonin following oral administration. For example assuming a volume of distribution of 1000 L we would predict that ingestion of 1000 μg melatonin would produce a blood level of approximately 1000 pg/ml; this is very close to the actual peak concentration of melatonin (1354 pg/ml) achieved 45 minutes after administration of 1000 μg melatonin to healthy volunteers (Dawson, et al., 1996). Using data from the other doses of melatonin administered in that study it is clear that there is a dose response relationship between the amount administered and the subsequent peak plasma levels (figure 1; (Dawson, et al., 1996)). Furthermore to increase the peak melatonin concentration by 5 pg/ml would require the administration of 3.5 μg (3,500,000 pg) melatonin.

Effect of "melatonin rich" food on plasma melatonin

Does the ingestion of melatonin rich foods lead to physiologically significant changes in plasma melatonin? Maldonaldo et al administered 660 ml beer containing 170 pg/ml melatonin to 4 men and 330 ml beer to 3 women aged between 20 and 30 years. The amount of melatonin ingested was therefore 112 ng for men and 56 ng for women. Blood was drawn before and 45 minutes

after consumption of the beer and melatonin was assayed by ELISA. (Maldonado, et al., 2009). While the actual average increase in plasma melatonin was not reported, based upon data presented in figure 1 of the paper, it can be estimated to be approximately 2.1 pg/ml.

In another study (Sae-Teaw, et al., 2013), blood was collected from subjects before breakfast and again after ingestion of either pineapple or orange juice or whole bananas. The estimated intake of melatonin was 302 ng, 150 ng and 1.7 ng for pineapple, orange and banana respectively. Plasma melatonin analysed by ELISA increased from 32 - 48 pg/ml, 60 minutes after eating the fruits to 145 – 151 pg/ml after 120 minutes and then decreased rapidly to be less than 50 pg/ml after 180 minutes.

Why are these results difficult to swallow? As previously mentioned, the bioavailability of melatonin is poor and the volume of distribution is approximately 1000 L. In both of the above studies the melatonin intake was between 50 and 300 ng. Following absorption and distribution of the oral melatonin, the maximum level that we might expect a 300 ng dose to achieve will be 300ng/1000 L, i.e., 0.3 pg/ml, well below that reported. Ingestion of approximately 100 times less melatonin in the bananas curiously resulted in similar plasma melatonin profiles. Moreover, the decrease in plasma melatonin levels between 120 and 180 minutes after ingestion of the fruit implies a shorter half life than has been previously reported. It is not known what was measured in the plasma melatonin assays, but it is reasonable to suggest that it was not melatonin.

Effect of “melatonin rich” food on melatonin metabolite excretion

Along with a potential increase in blood levels of melatonin following consumption of melatonin rich food, it might be expected that levels of melatonin metabolites would also increase. To address this, Oba et al (2008) altered the diet of subjects such that those on a control diet reduced

the estimated melatonin intake from 6 selected vegetables from 133 ng to 5 ng per day, while those on the intervention diet increased the estimated melatonin intake from 125 ng to 1288 ng per day (Oba, et al., 2008). The authors did not indicate when the vegetables were consumed or how they were prepared. Overnight first void concentrations of 6-sulphatoxy melatonin (aMT.6S) adjusted for creatinine were determined. After 65 days aMT.6S decreased by 4.7 ng/mg creatinine in the control group ($P < 0.05$) and increased by 1.5 ng/mg creatinine ($P > 0.05$) in the intervention group. To put this in perspective the following calculations can be made. The excretion of creatinine in human urine is approximately 24 mg/kg/24h (Greenblatt, et al., 1976), so subjects weighing 50kg will excrete 400 mg creatinine overnight. Therefore it can be estimated that the controls in the Oba et al (2008) study decreased their melatonin intake by 128 ng per day, but decreased aMT.6S excretion by 1880 ng, whereas the intervention group increased their melatonin intake by 1163 ng per day and yet only increased aMT.6S by 600 ng. Nevertheless the authors concluded that daily consumption of a high amount of selected vegetables increased 6-suphatoxymelatonin concentration in first void morning urine.

Jerte Valley cherries are reported to be rich in melatonin. When middle aged and elderly subjects consumed 200g of various cherry cultivars twice a day as lunch and dinner deserts the aMT.6S content of overnight first void urine was increased by approximately 20 – 80% over baseline depending upon the cultivar (Garrido, et al., 2010). If we presume that the average excretion of aMT.6S is 30 μg per day (Fellenberg, et al., 1980; Lane & Moss, 1985), this represents an increased excretion of 6 – 24 μg aMT.6S per day. The cherries used in this study are reported to have a concentration of 0 - 22.4 ng/100g fresh weight (González-Gómez, et al., 2009), which represented a daily intake of only 0 to 90 ng melatonin.

Studies using similar protocols have been published for a Jerte Valley cherry based product, plums and grape juice consumed by young, middle aged and elderly subjects. Consumption of the cherry product resulted in 1.5-, 2.4- and 2.5-fold increases in overnight first void aMT.6S excretion compared to baseline (Garrido, et al., 2013). Consumption of 195 g plums twice a day for 5 days increased overnight excretion of aMT.6S by 2-, 3- and 2-fold in young middle-aged and elderly participants respectively despite the fact that melatonin "was not quantifiable in the plums" (Gonzalez-Flores, et al., 2011). Grape juice (200 ml per day for 5 days) increased excretion of aMT.6S by 3-, 3- and 4-fold in the young, middle aged and elderly subjects respectively (Gonzalez-Flores, et al., 2012). The amount of melatonin ingested was not reported. Montmorency cherry juice concentrate or placebo of a fruit cordial containing less than 5% fruit were ingested by subjects within 30 of awakening and 30 minutes before the evening meal for 7 days (Howatson, et al., 2012). Serial urine collections were made over 48 hour periods for the interventions and aMT.6S excretion measured. Subjects drinking the cherry concentrate excreted approximately 4 μg aMT.6S compared to their baseline while the placebo group did not change. Based upon an independent analysis of the melatonin content, the concentrate apparently contained 1.42 μg/ml, which equated with a daily intake of approximately 85μg/day.

Finally in a follow-up study of the effects of their previous study on melatonin in fruit, urinary aMT.6S excretion was determined following ingestion of juice or pulp from 1 kg pineapple or orange and 190 g banana after an evening meal between 1800h and 1830h (Johns, et al., 2012). The total urine output from 1900h to 0700h was collected at baseline and the night of fruit consumption. Excretion of aMT.6S increased by 180%, 49% and 266% after consumption of pineapple, orange and banana respectively. This equates to increases of approximately 54 μg,

14.7 μg and 79.8 μg aMT.6S following the estimated intake of melatonin of 302 ng, 150 ng and 1.7 ng for pineapple, orange and banana respectively.

Why are these results difficult to swallow? The study by Oba et al (2008) seriously overstated the outcome of their intervention; when the authors analysed the positive intervention group in isolation, increasing the consumption of melatonin 10 fold by ingestion of melatonin rich foods did not significantly increase aMT.6S excretion (Oba, et al., 2008). The studies by the University of Extramadura group (Garrido, et al., 2013; Garrido, et al., 2010; Gonzalez-Flores, et al., 2012; Gonzalez-Flores, et al., 2011) on the effects of cherries, plums and grapes on melatonin metabolite excretion reported increases ranging from 6 to 150 μg per day. In other words ingestion of melatonin in the order of 100 ng resulted in 60 to 1500 fold increases in metabolite excretion in the overnight first void urine. The apparent many fold increases in metabolite are probably even larger since the foods were consumed during the afternoon as well as evening and the daytime ingestion would not have contributed to the observed overnight metabolite levels. Similar assessments can be for the Montmorency cherry (Howatson, et al., 2012) and pineapple, orange and banana studies (Johns, et al., 2012). The levels of metabolite excreted do not agree with the actual melatonin intake. It should be pointed out also that the concentration of melatonin reported in Montmorency cherry juice (Howatson, et al., 2012) is extraordinarily high, being approximately 10,000 times higher than previously reported in *any* food.

The use of urinary excretion of melatonin to support the potential utility of melatonin rich food for improving health is in itself curious. There is no question that many of those promoting the health benefits of consuming melatonin rich foods propose that the benefits arise from the anti-oxidant properties of the melatonin. For example in a recent review it was stated that “As an

antioxidant, the high levels of melatonin in plants are also beneficial to animals including humans, who consume them. Throughout the world, billions of people depend on these products as a major food source so the potentially beneficial effects of melatonin consumed in these products is obvious" (Tan, et al., 2012). By measuring aMT.6S excretion the researchers are in fact determining the amount of melatonin that has *escaped* oxidation, and instead they are measuring melatonin metabolised by CPY1A2. It would be more useful to measure the levels of the oxidative products of melatonin such as N1-acetyl-N2-formyl-5-methoxykynurenamine and N1-acetyl-5-methoxykynurenamine and/or their further metabolites or conjugates. However, as previously mentioned, these metabolites have not been detected even after ingestion of 10 mg melatonin (Li, et al., 2013).

What are the possible explanations for the detection of melatonin and its metabolites?

There are several possibilities that might explain the results that have been reported regarding the ingestion of melatonin rich foods. The foods may contain precursors of melatonin such as tryptophan or 5-hydroxy tryptophan which are then converted to melatonin. In a study involving the intra venous infusion of 5 grams of tryptophan over 40 minutes during daytime, the plasma melatonin concentration increased to 290 pg/ml, 60 minutes after the start of the infusion and then decreased to the expected low daytime levels after 5 hours (Heuther, et al., 1992). In another study, subjects ingested 6 grams of tryptophan and plasma melatonin levels increased approximately 12 pg/ml after five hours (Christofides, et al., 2006). In the case of oral administration of 600 – 800 mg 5-hydroxy tryptophan, plasma melatonin levels did not increase above the detection limit of 15.6 pg/ml (Cavallo, et al., 1987). It is thus unlikely that melatonin precursors are responsible for the reported melatonin increases.

The melatonin rich foods may contain CYP1A2 inhibitors which delay/prevent the metabolism of endogenous melatonin, leading to elevated plasma melatonin. It is known for example that certain foods, like carrots, celeriac, celery, coriander, cumin, grapefruit juice, parsnip and parsley may inhibit CYP1A2 (Perera, et al., 2012). The opposite argument would be required for studies that resulted in the many fold increases in melatonin metabolite excretion following ingestion of melatonin rich foods. While I am not aware of studies addressing the effects of the melatonin rich foods on CYP1A2 activity, if they did have significant activity, the overall health consequences for people taking drugs that are metabolised by CYP1A2 would be of major concern.

Finally the assays used to measure the plasma melatonin and urinary aMT.6S levels may not be specific for their target analytes and instead they may be detecting other interfering compounds that are perhaps related indoles, for example “melatonin isomers” (Gomez, et al., 2013; Kocadagli, et al., 2014) or unrelated compounds.

Conclusion

Melatonin is a remarkable compound that is found across plant and animals species in ng/g concentrations or lower. By virtue of studies in animals demonstrating anti-oxidant properties of endogenous and exogenous melatonin, a view has emerged that the melatonin in plants used as food may have beneficial health effects in humans. Studies reporting the appearance of melatonin in blood and its metabolites in urine following ingestion of melatonin rich foods appear to be flawed. The known pharmacokinetic profile of melatonin following its oral administration do not support the large changes that have been reported in the literature, which in some cases represent 5 fold increases in metabolite excretion over the endogenous production.

While there may be health benefits of ingestion of certain foods it is difficult to accept that they are due to their melatonin content.

References

Badria, F. A. (2002). Melatonin, serotonin, and tryptamine in some egyptian food and medicinal plants. *J. Med. Food,* **5**; 153-157.

Boden, M. J., Varcoe, T. J., & Kennaway, D. J. (2013). Circadian regulation of reproduction: From gamete to offspring. *Prog. Biophys. Mol. Biol.,* **113**; 387-397.

Cavallo, A., Richards, G. E., Meyer, W. J., & Waldrop, R. D. (1987). Evaluation of 5-hydroxytryptophan administration as a test of pineal function in humans. *Horm. Res.,* **27**; 69-73.

Christofides, J., Bridel, M., Egerton, M., Mackay, G. M., Forrest, C. M., Stoy, N., Darlington, L. G., & Stone, T. W. (2006). Blood 5-hydroxytryptamine, 5-hydroxyindoleacetic acid and melatonin levels in patients with either Huntington's disease or chronic brain injury. *J. Neurochem.,* **97**; 1078-1088.

Dardente, H. (2012). Melatonin-dependent timing of seasonal reproduction by the pars tuberalis: Pivotal roles for long daylengths and thyroid hormones. *J. Neuroendo.,* **24**; 249-266.

Dawson, D., Gibbon, S., & Singh, P. (1996). The hypothermic effect of melatonin on core body temperature: Is more better? *J. Pineal Res.,* **20**; 192-197.

DeMuro, R. L., Nafziger, A. N., Blask, D. E., Menhinick, A. M., & Bertino, J. S., Jr. (2000). The absolute bioavailability of oral melatonin. *J. Clin. Pharmacol.,* **40**; 781-784.

Dubbels, R., Reiter, R. J., Klenke, E., Goebel, A., Schnakenberg, E., Ehlers, C., Schiwara, H. W., & Schloot, W. (1995). Melatonin in edible plants identified by radioimmunoassay and by high performance liquid chromatography-mass spectrometry. *J. Pineal Res.,* **18**; 28-31.

Dubocovich, M. L., Delagrange, P., Krause, D. N., Sugden, D., Cardinali, D. P., & Olcese, J. (2010). International Union of Basic and Clinical Pharmacology. LXXV. Nomenclature, classification, and pharmacology of G protein-coupled melatonin receptors. *Pharmacol. Rev.,* **62**; 343-380.

Fellenberg, A. J., Phillipou, G., & Seamark, R. F. (1980). Measurement of urinary production rates of melatonin as an index of human pineal function. *Endocr.Res Commun.,* **7**; 167-175.

Fernández-Pachón, M. S., Medina, S., Herrero-Martín, G., Cerrillo, I., Berná, G., Escudero-López, B., Ferreres, F., Martín, F., García-Parrilla, M. C., & Gil-Izquierdo, A. (2014). Alcoholic fermentation induces melatonin synthesis in orange juice. *J. Pineal Res.,* **56**; 31-38.

Fourtillan, J. B., Brisson, A. M., Gobin, P., Ingrand, I., Decourt, J. P., & Girault, J. (2000). Bioavailability of melatonin in humans after day-time administration of D7 melatonin. *Biopharm. Drug Dispos.,* **21**; 15-22.

Garcia-Moreno, H., Calvo, J. R., & Maldonado, M. D. (2013). High levels of melatonin generated during the brewing process. *J. Pineal Res.,* **55**; 26-30.

Garrido, M., Gonzalez-Gomez, D., Lozano, M., Barriga, C., Paredes, S. D., & Moratinos, A. R. (2013). A jerte valley cherry product provides beneficial effects on sleep quality. Influence on aging. *J. Nutr. Health Aging,* **17**; 553-560.

Garrido, M., Paredes, S. D., Cubero, J., Lozano, M., Toribio-Delgado, A. F., Munoz, J. L., Reiter, R. J., Barriga, C., & Rodriguez, A. B. (2010). Jerte Valley cherry-enriched diets improve nocturnal rest and increase 6-sulfatoxymelatonin and total antioxidant capacity in the urine of middle-aged and elderly humans. *J. Gerontol. A Biol. Sci. Med. Sci.,* **65**; 909-914.

Gomez, F. J. V., Hernández, I. G., Martinez, L. D., Silva, M. F., & Cerutti, S. (2013). Analytical tools for elucidating the biological role of melatonin in plants by LC-MS/MS. *Electrophoresis,* **34**; 1749-1756.

Gonzalez-Flores, D., Gamero, E., Garrido, M., Ramirez, R., Moreno, D., Delgado, J., Valdes, E., Barriga, C., Rodriguez, A. B., & Paredes, S. D. (2012). Urinary 6-sulfatoxymelatonin and total antioxidant capacity increase after the intake of a grape juice cv. Tempranillo stabilized with HHP. *Food Funct.,* **3**; 34-39.

Gonzalez-Flores, D., Velardo, B., Garrido, M., Gonzalez-Gomez, D., Lozano, M., Ayuso, M. C., Barriga, C., Paredes, S. D., & Rodriguez, A. B. (2011). Ingestion of Japanese plums (Prunus salicina Lindl. cv. Crimson Globe) increases the urinary 6-sulfatoxymelatonin and total antioxidant capacity levels in young, middle-aged and elderly humans:

Nutritional and functional characterization of their content. *J. Food Nutr. Res.,* **50**; 229-236.

González-Gómez, D., Lozano, M., Fernández-León, M. F., Ayuso, M. C., Bernalte, M. J., & Rodríguez, A. B. (2009). Detection and quantification of melatonin and serotonin in eight Sweet Cherry cultivars (Prunus avium L.). *Eur Food Res Technol,* **229**; 223-229.

Greenblatt, D. J., Ransil, B. J., Harmatz, J. S., Smith, T. W., Duhme, D. W., & Koch-Weser, J. (1976). Variability of 24-hour urinary creatinine excretion by normal subjects. *J. Clin. Pharmacol.,* **16**; 321-328.

Hattori, A., Migitaka, H., Iigo, M., Itoh, M., Yamamoto, K., Ohtani Kaneko, R., Hara, M., Suzuki, T., & Reiter, R. J. (1995). Identification of melatonin in plants and its effects on plasma melatonin levels and binding to melatonin receptors in vertebrates. *Biochem. Mol. Biol. Int.,* **35**; 627-634.

Heuther, G., Hajak, G., Reimer, A., Poeggeler, B., Blomer, M., Rodenbeck, A., & Ruther, E. (1992). The metabolic fate of infused L-tryptophan in men: possible clinical implications of the accumulation of circulating tryptophan and tryptophan metabolites. *Psychopharmacology (Berl),* **109**; 422-432.

Hirata, F., Hayaishi, O., Tokuyama, T., & Seno, S. (1974). In vitro and in vivo formation of two new metabolites of melatonin. *J. Biol. Chem.,* **249**; 1311-1313.

Howatson, G., Bell, P., Tallent, J., Middleton, B., McHugh, M., & Ellis, J. (2012). Effect of tart cherry juice (Prunus cerasus) on melatonin levels and enhanced sleep quality. *Eur J Nutr,* **51**; 909-916.

Huang, X., & Mazza, G. (2011). Application of LC and LC-MS to the analysis of melatonin and serotonin in edible plants. *Crit. Rev. Food Sci. Nutr.,* **51**; 269-284.

Johns, N. P., Johns, J., Porasuphatana, S., Plaimee, P., & Sae-Teaw, M. (2012). Dietary intake of melatonin from tropical fruit altered urinary excretion of 6-sulfatoxymelatonin in healthy volunteers. *J. Agric. Food Chem.,* **61**; 913-919.

Jones, R. L., McGeer, P. L., & Greiner, A. C. (1969). Metabolism of exogenous melatonin in schizophrenic and non- schizophrenic volunteers. *Clinica Chimica Acta,* **26**; 281-285.

Keijzer, H., Smits, M. G., Duffy, J. F., & Curfs, L. M. G. (2014). Why the dim light melatonin onset (DLMO) should be measured before treatment of patients with circadian rhythm sleep disorders. *Sleep Med. Rev.,* **18**; 333-339.

Kocadagli, T., Yılmaz, C., & Gökmen, V. (2014). Determination of melatonin and its isomer in foods by liquid chromatography tandem mass spectrometry. *Food Chem.,* **153**; 151-156.

Lane, E. A., & Moss, H. B. (1985). Pharmacokinetics of melatonin in man: first pass hepatic metabolism. *J. Clin. Endocrinol. Metab.,* **61**; 1214-1216.

Li, C. Y., Li, G. M., Tan, D. X., Li, F., & Ma, X. C. (2013). A novel enzyme-dependent melatonin metabolite in humans. *J. Pineal Res.,* **54**; 100-106.

Ma, X., Idle, J. R., Krausz, K. W., & Gonzalez, F. J. (2005). Metabolism of melatonin by human cytochromes p450. *Drug Metab Dispos.,* **33**; 489-494.

Maldonado, M. D., Moreno, H., & Calvo, J. R. (2009). Melatonin present in beer contributes to increase the levels of melatonin and antioxidant capacity of the human serum. *Clin. Nutr.,* **28**; 188-191.

Oba, S., Nakamura, K., Sahashi, Y., Hattori, A., & Nagata, C. (2008). Consumption of vegetables alters morning urinary 6-sulfatoxymelatonin concentration. *J. Pineal Res.,* **45**; 17-23.

Paredes, S. D., Korkmaz, A., Manchester, L. C., Tan, D.-X., & Reiter, R. J. (2009). Phytomelatonin: a review. *J. Exp. Bot.,* **60**; 57-69.

Peng, H. T., Bouak, F., Vartanian, O., & Cheung, B. (2013). A physiologically based pharmacokinetics model for melatonin—Effects of light and routes of administration. *Int. J. Pharm.,* **458**; 156-168.

Perera, V., Gross, A. S., & McLachlan, A. J. (2012). Influence of environmental and genetic factors on CYP1A2 activity in individuals of south asian and european ancestry. *Clin. Pharmacol. Ther.,* **92**; 511-519.

Pierpaoli, W., & Regelson, W. (1995). *The melatonin miracle*. New York: Simon and Schuster.

Reiter, R. J., Manchester, L. C., & Tan, D. X. (2005). Melatonin in walnuts: influence on levels of melatonin and total antioxidant capacity of blood. *Nutrition,* **21**; 920-924.

Reiter, R. J., Tan, D. X., & Fuentes-Broto, L. (2010). Melatonin: a multitasking molecule. *Prog. Brain Res.,* **181**; 127-151.

Sae-Teaw, M., Johns, J., Johns, N. P., & Subongkot, S. (2013). Serum melatonin levels and antioxidant capacities after consumption of pineapple, orange, or banana by healthy male volunteers. *J. Pineal Res.,* **55**; 58-64.

Tan, D.-X., Hardeland, R., Manchester, L. C., Korkmaz, A., Ma, S., Rosales-Corral, S., & Reiter, R. J. (2012). Functional roles of melatonin in plants, and perspectives in nutritional and agricultural science. *J. Exp. Bot.,* **63**; 577-597.

Table 1. Concentration of melatonin in various foods. Data are shown as pg/g or pg/ml.

Food	Melatonin concentration	Reference
Walnut	137 pg/g	(Kocadagli, et al., 2014)
Walnut	3400 pg/g	(Reiter, et al., 2005)
Orange juice	2500 pg/ml	(Fernández-Pachón, et al., 2014)
Orange	150 pg/g	(Johns, et al., 2012)
Banana	8.9 pg/g	(Johns, et al., 2012)
Banana	Not Detected	(Huang & Mazza, 2011)
Banana	655 pg/g	(Badria, 2002)
Banana	470 pg/g	(Dubbels, et al., 1995)
Pineapple	302 pg/g	(Johns, et al., 2012)
Pineapple	280 pg/g	(Badria, 2002)
Pineapple	40 pg/ml	(Hattori, et al., 1995)
Sour cherry concentrate	<17.2 pg/ml	(Kocadagli, et al., 2014)
Sour cherry concentrate	1,420,000 pg/ml	(Howatson, et al., 2012)
Merlot	Not Detected	(Gomez, et al., 2013)
Mid strength beer	113 pg/ml	(Garcia-Moreno, et al., 2013)

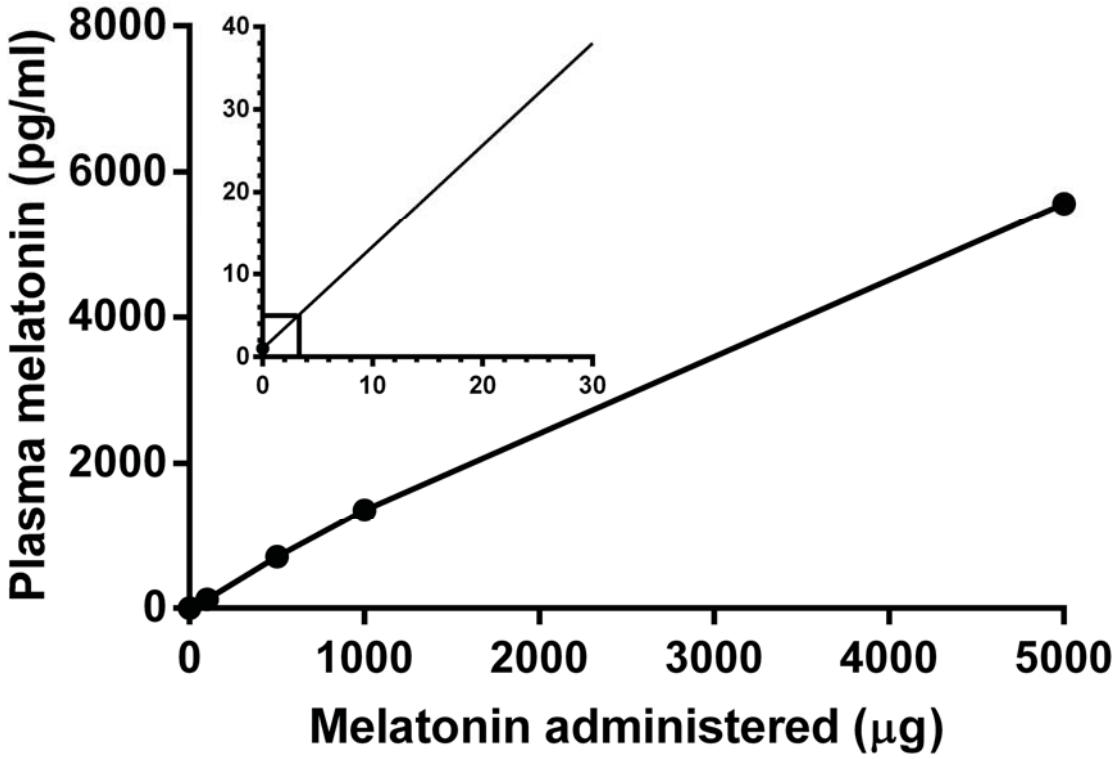

Figure 1 Peak plasma melatonin concentration following the oral ingestion of 100 μg, 500 μg, 1000 μg and 5000 μg melatonin using data from (Dawson, et al., 1996). The insert shows the data between 0 and 30 μg melatonin. From this it can be estimated that to achieve a 5 pg/ml increase in melatonin, approximately 3.5 μg would need to be consumed.

