# National Journal of Physiology, Pharmacy and Pharmacology

## RESEARCH ARTICLE

## Effect of aircraft fuel tank maintenance on salivary cortisol secretion

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**Received:** May 09, 2017; **Accepted:** Jun 21, 2017

#### **ABSTRACT**

**Background:** Chemical exposure of human by inhalation or skin exposure to confined space is two stressful events that may cause trauma. In case of aircraft maintenance, workers have to withstand the confined space with poorly-ventilated environment, where they are exposed to toxic hazardous vapor coming from the jet fuel which mainly is kerosene based (>98%). The use of salivary cortisol is very well established stress biomarker and hence can be used as a tool to measure stress level. **Aims and Objectives:** This study sought to find out the effect of exposure to aircraft fuel tank during maintenance work on the salivary cortisol secretion. **Materials and Methods:** A total of 95 healthy volunteers (67 males, 28 females) aged between 17 and 25 years old took part in this study. Saliva samples were collected before and after fuel tank entry for 15 min. Cortisol concentration was determined by enzyme-linked immune sorbent assay (ELISA) Salivary cortisol kit by Salimetrics. **Results:** The cortisol level increased significantly (P=0.001) after exposure to kerosene vapor and confined space. The means of cortisol levels before and after fuel tank entry were 0.28 μg/dL and 0.35 μg/dL, respectively. The hostile characteristic of a confined space is an excellent location to form hazardous concentrations since kerosene contains chemicals with a vapor density greater than air. **Conclusion:** This study indicated that exposure to kerosene vapor and confined space will cause a physiological stress and trigger a physiological response in the body of the exposed person.

KEY WORDS: Aircraft Fuel Tank, Confined Space, Jet Fuel, Kerosene Vapor, Salivary Cortisol and Stress

#### INTRODUCTION

The aircraft fuel tank is known as a hazardous workspace which exposes the workers with various hazardous components such as risk of fire and explosion, toxic vapor from the jet fuel and other irritating chemicals, confined space and oxygen deficiency. According to Boeing, the most common hazard of an aircraft fuel tank is the jet fuel

Access this article online		
Website: www.njppp.com	Quick Response code	
<b>DOI:</b> 10.5455/njppp.2017.7.0515821062017		

itself and more importantly, the most commonly used jet fuels in commercial flights are Jet A and Jet A-1 which are the kerosene-type jet fuel. [1] Kerosene, on the other hand, is known as a toxic compound which according to the Health Protection Agency United Kingdom, can cause adverse health effects and symptoms if inhaled in large quantities. In fact, long-term exposure to jet fuel vapor through activities such as fueling and maintaining large numbers of aircrafts has been associated with deficits in central nervous system function which includes fatigue, neurobehavioral changes, psychiatric disorders, and abnormal electroencephalogram. [2,3]

The confined nature of the fuel tank itself may pose several threats to the maintenance workers. An aircraft fuel tank is normally only accessible through a small opening that allows only one small-to-medium-sized person to enter horizontally.

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The interior structures are further divided into a few rib sections which are interconnected through access holes that can only accommodate the worker's head, shoulder and trunk, leaving the legs outside. These small opening and access holes, therefore, inhibit proper air flow in the fuel tank and resulting in inadequate ventilation associated with oxygen deficiency. At oxygen-deficient levels, a person might show symptoms of oxygen deprivation such as headache, vomiting, drowsiness, and slurred speech and in worst case scenario death. Besides, entry into a confined enclosed space may trigger sudden onset of claustrophobia resulting in panic and various physiological malfunction.<sup>[4]</sup>

The hypothalamic-pituitary-adrenal axis is the major neuroendocrine stress pathway within a human body which will be triggered in both psychological and physiological stress. Triggering this pathway will lead to the secretion of cortisol which consequently affects the metabolic, cardiovascular and central nervous systems.[5] In fact, the change of cortisol level in the body has been used as an important biomarker for anxiety disorders such as in the event of depression and posttraumatic stress. [6] Maruyama et al. (2012) found a significant correlation between salivary cortisol and the trier social stress tests (resembling psychosocial stress) in healthy female volunteers.[7] Therefore, the use of salivary cortisol as stress biomarker has nowadays been widely accepted because it is noninvasive and stress-free.[8] Despite the hostile environment of the aircraft fuel tank, the maintenance and repair works have to be done on a regular basis after a certain number of flight hours. In fact, this is the most crucial component of the commercial aviation to make sure that a plane is in the best condition before taking off to avoid any unfortunate incidences along the journey. Therefore, this project sought to study the association of exposure to aircraft fuel tank during maintenance and repair work with the salivary cortisol secretion. A simulated aircraft fuel tank was prepared, and a group of students of the Malaysian Institute of Aviation Technology (MIAT) made the fuel tank entry. Saliva samples were collected before and after fuel tank entry and further subjected to cortisol measurement. The data on the level of cortisol of this group will give us an insight on the stress level experienced by the aircraft maintenance workers due to exposure to hazardous environment in the fuel tank.

#### MATERIALS AND METHODS

Research was conducted at the hangar of Universiti Kuala Lumpur MIAT, Dengkil Selangor involving 95 healthy volunteers (67 males, 28 females) aged between 17 and 25 years old. Verbal and written consents were obtained and basic medical checkup (blood pressure, body temperature, and electrocardiogram) was done before fuel tank entry to make sure that respondents were in good health before entering the aircraft fuel tank. Respondents were given

15 min each to do the inspection and maintenance tasks in the tank. Since cortisol follows a circadian rhythm with peak level at 8 am and is nadir in the late evening, tank entry and sample collection were scheduled between 8 and 11 am to standardize and minimize the cortisol oscillation. [9,10]

### **Fuel Tank Preparation and Entry**

The procedures for fuel tank preparation and entry were done according to the rules and regulations outlined in the Boeing 737-600/700/800/900 maintenance manual which involved purging and biocide treatment of the fuel tank. Proper attire for fuel tank entry was strictly followed as given in the manual. [11] Based on Boeing 737-300/400/500 maintenance manual, the time allowed for each worker to do the aircraft fuel tank maintenance and inspection is between 15 min and 8 h. [11] However, to standardize the exposure time in this study, we used 15 min for each respondent to be in aircraft fuel tank to do the inspection.

#### **Saliva Sampling**

Saliva was collected in dried and sterilized 50 ml centrifuge tubes before and after fuel tank entry. Protocol for saliva sampling was adopted from Chiappin et al. (2007), with minor modification. One piece of 4 cm × 4 cm Parafilm was given for the respondents to chew to stimulate saliva production. Whole saliva sample with visible blood contamination would be excluded. As much as 5 ml of whole saliva would be collected, and immediately 1.5 ml of saliva would be transferred into a dried and sterilized 1.5 ml centrifuge tube and stored in ice. All the samples were later stored at -20°C until determination of cortisol level was done.

#### **Salivary Cortisol Level Measurement**

Cortisol concentration was determined by ELISA salivary cortisol kit by Salimetrics<sup>®</sup>. All samples were treated following the protocol provided by the manufacturer. Samples of both before and after were tested in duplicate. Cortisol standards provided by the manufacturer were included in each run. ELISA plates were read with a chromate awareness technology plate reader using a 450 nm filter. The concentration of cortisol in each saliva sample was then calculated based on the procedure and protocol from the kit manual.

#### **Statistical Analysis**

All data were analyzed using SPSS version 17.0 for windows.

#### **RESULTS**

The cortisol level in respondents increased significantly (P = 0.001) after performing aircraft fuel tank maintenance

check. The means of cortisol level before and after fuel tank entry were 0.29  $\mu g/dL$  and 0.35  $\mu g/dL$ , respectively, as shown in Table 1 and Figure 1.

#### **DISCUSSION**

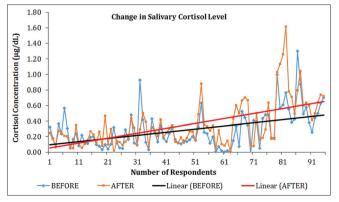
In aircraft fuel tank maintenance, the workers have to physically enter the tank which exposes them to the environmental hazards exist in the space. The main hazard of the aircraft fuel tank is eventually the fuel tank itself and the most commonly used jet fuel in commercial flights are Jet A and Jet A-1 which are kerosene-based (98+%).[1] Goodwin et al. demonstrated that kerosene aspiration resulted in severe and persistent intrapulmonary physiology shunting. bradycardia, hypoxemia, and hypotension in canine model.<sup>[14]</sup> The previous findings also reported two types of kerosene effects on the lung. The primary effects are atelectasis, increase of inflammatory cells, lipoid pneumonia, and loss of effectiveness of lung surfactant, [15] while the secondary effects are pneumothorax, pneumatocele or broncho plural fistula.[16] These effects are due to the low viscosity and surface tension of kerosene, which allow it to be aspirated into the lungs of person who is exposed.

The situation becomes more complicated when the kerosene vapor and confined space work together to induce the impact on human health and safety. The hostile characteristic of a confined space is an excellent location to form hazardous concentrations since kerosene contains chemicals with a vapor density greater than air. These heavier-than-air chemical vapors are particularly unstable/dangerous because concentration levels are heaviest at the lowest point and

**Table 1:** The difference in salivary cortisol levels before and after the aircraft fuel tank entry

Time	n	Mean±SD
Before	95	0.29±0.24
After	95	0.35±0.30

SD: Standard deviation



**Figure 1:** The difference in salivary cortisol levels before and after the aircraft fuel tank entry

gradually diminish as the height increases. Unfortunately, this poses two problems. First is when personnel enters into a confined space at a level that generally exposes the worker to a higher concentration of toxic materials. Entry without proper respiratory protection or even sudden onset of claustrophobia may result in collapse. The worker then becomes exposed to even more concentrated level of contamination when falling to the ground. The second problem arises when atmosphere testing was conducted before entry, but measurements were not taken at the lowest level of the confined space. Therefore, incomplete readings may be taken if a probe dropped from the top of the tank did not reach the bottom.<sup>[17]</sup>

Cortisol is the major glucocorticoid produced in the adrenal cortex. Cortisol production has a circadian rhythm.<sup>[18]</sup> The level peaks in the early morning and drops to the lowest concentration at night.[19] However, the level rises independently of circadian rhythm in response to stress.[20] In blood, only about 5-10% of cortisol is unbound or biologically active, the remaining cortisol is bound to serum proteins.[21] The unbound serum cortisol enters the saliva via intracellular mechanisms and remains unbound to protein.[22] Salivary cortisol levels are unaffected by salivary flow rate and are relatively resistant to degradation from enzymes or freeze-thaw cycles.[22,23] Studies consistently reported high correlations between serum and saliva cortisol. indicating that salivary cortisol levels reliably estimate serum cortisol levels. [24-26] Like other glucocorticoids, once cortisol is released, it will interact with the glucocorticoid and mineralocorticoid receptors and acts throughout the body mediating alterations in various processes such as the immune reaction, metabolic regulation, and inflammatory reaction.[27]

Body uses the three steps adaptation to cope with stress. The first stage is activation of adrenal gland by the alarm reaction. On encountering a stressor, body reacts with "fight-or-flight" response through activation of sympathetic nervous system. In this stage, hormone such as cortisol and adrenalin is released into the bloodstream. The second stage is resistance where readjustment occurs. Parasympathetic nervous system returns many physiological functions to normal levels when body focuses on resources against the stressor. In addition, blood glucose level remains high with cortisol and adrenalin continue to circulate at elevated levels, but the outward appearance of the human seems normal. Eventually, the heart rate, blood pressure, and breathing will increase and at this stage, the body remains on red alert. The third stage is exhaustion if stressor continues beyond body's capacity; organism exhausts of resources and becomes susceptible to disease and death.[28]

Glucocorticoids suppress the immune systems, it is reasonable to expect that chronic stress may lead to an increased risk of illness. Indeed, it appears that there is an association between chronic stress and the susceptibility to disease.

Furthermore, cortisol level can increase in proportion to the severity of stress to as high as 6 times basal levels. This chronically high cortisol secretion may cause atrophy of the hippocampus (a brain region involved in memory and other function), possibly due to inhibition of neurogenesis. The high secretion of corticotropin-releasing hormone (CRH) from the hypothalamus, which drives the pituitary-adrenal axis, may also affect certain region of the brain, for example; there are CRH receptors in the amygdala, which involves in fearful memories. Therefore, scientists believe that high cortisol and or high CRH may act on the brain in chronic stress to contribute to anxiety and depression.[29] Therefore, glucocorticoids increase stress responses in brain and serve to regulate or contain the responses, thus helping recovery and behavioral alteration. Containment and regulation are important as a number of sites in the brain can be negatively affected by increased and prolonged exposure to glucocorticoids. Although moderate levels of these hormones are responsible for normal cell functioning, glucocorticoids must be maintained at appropriate levels to preserve the homeostatic function of body systems.<sup>[27]</sup>

Our study found a significant increase in the levels of salivary cortisol in respondents exposed to the aircraft fuel tank. This shows that exposure to kerosene vapor and confined space in as minimum as 15 min in the aircraft fuel tank time would lead to physiological stress reflected by an increase in the salivary cortisol. Based on our observations, the respondents were looking fine after coming out from the tank. This finding supports the study by selye that the organism may have seems normal, but the cortisol will continue to circulate at elevated level on encountering a physiological stress before an adaptation mechanism is completed. [28]

#### **CONCLUSION**

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The study sought to find the correlation between the exposure to aircraft fuel tank with salivary cortisol secretion among the aircraft maintenance workers. There was a significant increase in salivary cortisol level among the respondents following fuel tank entry. Thus, this indicates that exposure to kerosene vapor and confined space will cause a physiological stress and trigger a physiological response in the body of the exposed person. One of the physiological responses involved is increased in the level of cortisol. However, this is an acute effect of exposure to aircraft fuel tank. Further study is required on the subchronic and chronic effects of the exposure involving other physiological parameters. The impacts of chronic exposures to aircraft fuel tank on health, for e.g., on the pulmonary, cardiovascular, and nervous systems have been studied by other research groups. The information on how exposure to aircraft fuel tank poses stress to the workers is important in highlighting the safety issues at the airplane maintenance workplace to avoid incidences such as collapse and work-related accidents due to nervous system dysfunction. Therefore, the finding of this study will urge the authorized body to enforce the use of personal protective equipment among the aircraft maintenance workers at all time while performing their job and to ensure that all the safety procedures are strictly adhered to. Continuous monitoring of the employees' health is also highly suggestive as a means of ensuring the worker's health and safety at the workplace.

#### REFERENCES

- 1. Feeler RA. Safety specialists recommend precautions for work in aircraft fuel tank. Flight Saf Found Aviat Mech Bull. 1999;47(6):1-6.
- 2. Knave B, Mindus P, Struwe G. Neurasthenic symptoms in workers occupationally exposed to jet fuel. Acta Psychiatr Scand. 1979;60(1):39-49.
- 3. Lin B, Ritchie GD, Rossi J 3<sup>rd</sup>, Pancrazio JJ. Identification of target genes responsive to JP-8 exposure in the rat central nervous system. Toxicol Ind Health. 2001;17(5-10):262-9.
- 4. Sorrels D. Hazards of Airplanes Fuel Tank Entry. Safety and Health, Boeing Commercial Airplane Group. Available from: http://www.boeing.com/commercial/aeromagazine/aero\_04/textonly/s01txt.html.
- 5. Dedovic K, Duchesne A, Andrews J, Engert V, Pruessner JC. The brain and the stress axis: The neural correlates of cortisol regulation in response to stress. Neuroimage. 2009;47(3):864-71.
- 6. Sriram K, Rodriguez-Fernandez M, Doyle FJ. Modelling cortisol dynamic in the neuro-endocrine axis distinguishes normal, depression, and post-traumatic stress disorder (PTSD) in human. PLoS Comput Biol. 2012;8(2):1-15.
- Maruyama Y, Kawano A, Okamoto S, Ando T, Ishitobi Y, Tanaka Y, et al. Differences in salivary alpha-amylase and cortisol responsiveness following exposure to electrical stimulation versus the Trier Social Stress Tests. PLoS One. 2012;7(7):e39375.
- 8. Bozovic D, Racic M, Ivkovic N. Salivary cortisol levels as a biological marker of stress reaction. Med Arch. 2013;67(5):374-7.
- Bergendahl M, Vance ML, Iranmanesh A, Thorner MO, Veldhuis JD. Fasting as a metabolic stress paradigm selectively amplifies cortisol secretory burst mass and delays the time of maximal nyctohemeral cortisol concentrations in healthy men. J Clin Endocrinol Metab. 1996;81(2):692-9.
- 10. Lippi G, De Vita F, Salvagno GL, Gelati M, Montagnana M, Guidi GC. Measurement of morning saliva cortisol in athletes. Clin Biochem. 2009;42(9):904-6.
- 11. Boeing 737-300/400/500 Maintenance Manual; 28-10-00. Available from: http://www.academia.edu/8278925/Boeing\_737-300\_400\_500\_Aircraft\_Maintenance\_Manual.
- 12. Chiappin S, Antonelli G, Gatti R, De Palo EF. Saliva specimen: A new laboratory tool for diagnostic and basic investigation. Clin Chim Acta. 2007;383(1-2):30-40.
- 13. Bots CP, Brand HS, Veerman EC, van Amerongen BM, Nieuw Amerongen AV. Preferences and saliva stimulation of eight different chewing gums. Int Dent J. 2004;54(3):143-8.
- 14. Goodwin SR, Berman LS, Tabeling BB, Sundlof SF. Kerosene aspiration: Immediate and early pulmonary and cardiovascular effects. Vet Hum Toxicol. 1988;30(6):521-4.

- 15. Benois A, Petitjeans F, Raynaud L, Dardare E, Sergent H. Clinical and therapeutic aspects of childhood kerosene poisoning in Djibouti. Trop Doct. 2009;39(4):236-8.
- 16. Verma SK, Kapoor N, Bhaskar R, Upadhyay R. Pyopneumothorax following suicidal kerosene ingestion. BMJ Case Rep. 2012;2012. pii: Bcr2012007795.
- 17. Chilcott RP. Compendium of Chemical Hazards: Kerosene (Fuel Oil). Oxfordshire: H.P. Agency; 2006.
- 18. Dorn LD, Lucke JF, Loucks TL, Berga SL. Salivary cortisol reflects serum cortisol: Analysis of circadian profiles. Ann Clin Biochem. 2007;44:281-4.
- 19. Chernow B, Alexander HR, Smallridge RC, Thompson WR, Cook D, Beardsley D, et al. Hormonal responses to graded surgical stress. Arch Intern Med. 1987;147(7):1273-8.
- 20. Krieger DT. Rhythms of ACTH and corticosteroid secretion in health and disease, and their experimental modification. J Steroid Biochem. 1975;6(5):785-91.
- 21. Aardal E, Holm AC. Cortisol in saliva-reference ranges and relation to cortisol in serum. Eur J Clin Chem Clin Biochem. 1995;33(12):927-32.
- 22. Vining RF, McGinley RA. The measurement of hormones in saliva: Possibilities and pitfalls. J Steroid Biochem. 1987;27(1-3):81-94.
- Garde AH, Hansen AM. Long-term stability of salivary cortisol. Scand J Clin Lab Invest. 2005;65(5):433-6.

- Hiramatsu R. Direct assay of cortisol in human saliva by solid phase radioimmunoassay and its clinical applications. Clin Chim Acta. 1981:117(2):239-49.
- 25. Vining RF, McGinley RA, Maksvytis JJ, Ho KY. Salivary cortisol: A better measure of adrenal cortical function than serum cortisol. Ann Clin Biochem. 1983;20:329-35.
- 26. Francis SJ, Walker RF, Riad-Fahmy D, Hughes D, Murphy JF, Gray OP. Assessment of adrenocortical activity in term newborn infants using salivary cortisol determinations. J Pediatr. 1987;111(1):129-33.
- Franklin TB, Saab BJ, Mansuy IM. Neural mechanisms of stress resilience and vulnerability. Neuron. 2012;75(5): 747-61.
- 28. Selye H. The Stress of Live. New York: McGraw-Hill; 1987.
- 29. Fox. Human physiology. Endocrine Glands. 12<sup>th</sup> ed. New York: McGraw-Hill; 2012. p. 336.

**How to cite this article:** Latif NA, Mamat SNZ, Talib AHQA, Firdous J, Mokhtar AZ, Muhamad N. Effect of aircraft fuel tank maintenance on salivary cortisol secretion. Natl J Physiol Pharm Pharmacol 2017;7(9):886-890.

Source of Support: Nil, Conflict of Interest: None declared.