

## **Glutathione-s-transferase, reduced glutathione and oxidized glutathione: An adjuvant to general anesthesia with halothane or isoflurane**

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**Abstract:** Halothane and enflurane administration increased the plasma GSH-Px activity and reduced zinc levels. In addition, they lowered SOD and GSH-Px activities and trace element levels on erythrocytes. Isoflurane had no effect on plasma antioxidant enzymes, but, similar to the other, isoflurane decreased the plasma zinc levels, erythrocyte SOD and GSH-Px activities and trace element levels.

**Key words:** Glutathione-s-transferase, Halothane, Isoflurane

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### **I. Introduction**

Subclinical hepatic injuries after anesthesia continue to provoke interest particularly with newer, more sensitive methods of assessment such as measurement of plasma Glutathione-S transferase (GST), reduced glutathione, and oxidized glutathione concentration.

GST is rapidly released in circulation after hepatic damage and its short plasma half-life allow early detection of hepatic injury and its resolution. Hepatic blood flow usually decreases during regional and general anesthesia. Multiple factors are probably responsible including both direct and indirect effect of anesthetic agent<sup>1</sup>.

Reduced liver blood flow is said to be responsible for the observed increase in glutathione-s-transferase concentration after anesthesia<sup>2</sup>. Halothane cause hepatic blood flow to decrease in proportion to the depression of cardiac output. This reduced blood flow responsible for increase Glutathione concentration and hepatic cellular dysfunction includes minor transaminase elevation. Plasma glutathione-s-transferase, reduced glutathione and oxidized glutathione concentration measurement is sensitive and specific index of hepatocellular injury, GST concentration increase after anesthesia with most volatile anesthetic agent<sup>3</sup> (Darling et al, 2000). Plasma concentration of hepatic glutathione-s-transferase (GST) are a more sensitive measure of acute hepatic damage than aminotransferase activity<sup>4</sup>.

All volatile anesthetic agents reduce portal hepatic blood flow, this decrease is greatest with halothane and least with Isoflurane. Hussey<sup>5</sup> observed that small but significant increase in Glutathione-S-transferase concentration in patient receiving halothane or enflurane suggest an impairment of hepatocellular integrity following the administration of these anesthetics, in contrast, isoflurane anesthesia did not appear to be associated with this effect.

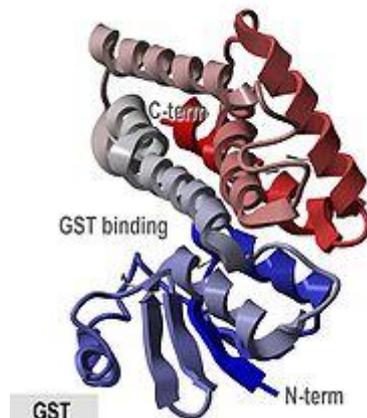
Allan et al<sup>6</sup> reported that in clinically identical situation anesthesia with halothane but not isoflurane lead to hepatocellular injury. However Rohm et al<sup>7</sup> observed that propofol have no influence on hepatocellular function during after surgery.

Isoflurane with its low blood gas coefficient and rapid elimination are thought to preclude prolonged elevated concentration of metabolite and resultant organ dysfunction. Standard biochemical test of liver function may have limited use in detection of minor degree of anesthesia related liver dysfunction. In contrast GST concentration in plasma provides a highly specific test of hepatocellular damage<sup>3</sup>.

Halothane<sup>8,9</sup> is oxidized in liver by a particular isozyme of cytochrome p-450 to its metabolite. Halothane causes hepatic blood flow to decrease in proportion to the depression of cardiac output. Hepatic artery vasospasm has been reported during halothane anesthesia. This reduced blood flow is responsible for observed increase in Glutathione-s-transferase concentration, hepatic cellular dysfunction and transaminase elevation.

In healthy cells and tissues, more than 90% of total glutathione pool is in the reduced form (GSH) and less than 10% exist in oxidized form (GSSG). An increased oxidized glutathione (GSSG) to reduced glutathione (GSH) ratio is considered to be indicative of oxidative stress. Thus we will measure GST concentration along with the reduced and oxidized glutathione concentration in patients undergoing general anesthesia with halothane or isoflurane to assess its effect on the hepatocellular integrity<sup>10</sup>.

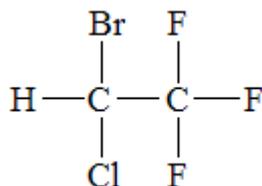
Mammalian cytosolic GSTs are dimeric both subunits being from the same class of GSTs, although not necessarily identical. The monomers are in the range of 22–29 kDa. They are active over a wide variety of substrates with considerable overlap.



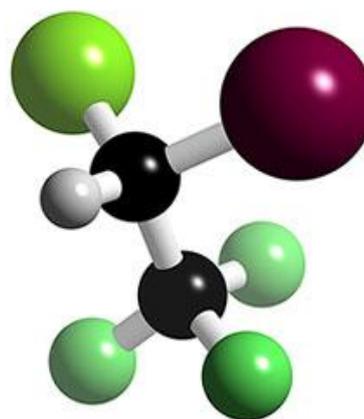
Glutathione-S-transferases are considered, among several others, to contribute to the phase II biotransformation of xenobiotics. Drugs, poisons, and other compounds not traditionally listed in either group are usually somewhat modified by the phase I and/or phase II mechanisms, and finally excreted from the body. GSTs contribute to this type of metabolism by conjugating these compounds (often electrophilic and somewhat lipophilic in nature) with reduced glutathione to facilitate dissolution in the aqueous cellular and extracellular media, and, from there, out of the body.

## II. Halothane

Halothane inhalational general anaesthetic. It is the only inhalational anaesthetic agent containing a bromine atom; there are several other halogenated anesthesia agents which lack the bromine atom and do contain the fluorine and chlorine atoms present in halothane. It is colourless and pleasant-smelling, but unstable in light. It is packaged in dark-coloured bottles and contains 0.01% thymol as a stabilising agent. Halothane has blood gas coefficient 2.3, hence medium range of solubility. It is first distributed to rich blood supply organ like brain, liver, kidney, then to muscle and fat with in 20 min. There is prolonged uptake of halothane by body as it is absorbed in the fat in sufficient amount. the solubility coefficient of fat is 60.

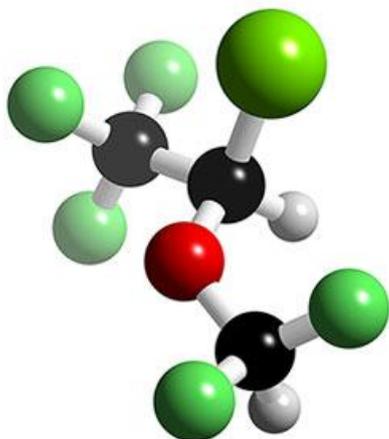


About 20% percent of inspired halothane is metabolized by liver microsomes and resulting product are excreted in urine. Volatile anesthetics especially halothane, can produce hepatic damage although the mechanism by which this rare but severe cause of fulminant hepatic failure occurs is unknown, prolonged administration of such anesthetics is a risk factor. Serum<sup>3</sup> or plasma concentration of alanine aminotransferase [ALT] and aspartate aminotransferase [AST] are commonly used to assess hepatic injury. However, because the aminotransferase are not specific to the liver and can be released under other condition, it is often difficult to ascribe small abnormalities in their plasma activities to hepatic damage. In addition, the aminotransferase, which are found mainly in periportal hepatocytes are relatively insensitive markers of damage to centilobular hepatocytes<sup>11</sup>, the cell most prone to damage by halothane, alcohol, toxin and hypoxia. Most of the volatile anaesthetics reduces hepatic blood flow, particularly halothane, halothane have marked effect in decreasing hepatic flow and inhibiting drug metabolism. Halothane has been linked to post operative liver dysfunction two syndrome are recognized, first is associated with transient rise in liver test, often after initial exposure. The second thought to occur after repeated exposure and has an immune mechanism with the development of fulminant hepatic failure. Chance of an immune reaction to a volatile agent occurring

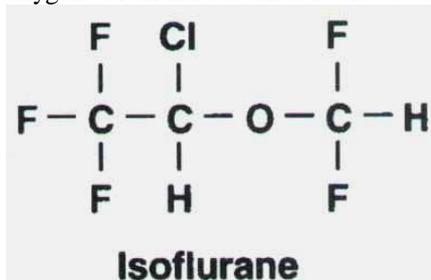


is thought to relate to the amount it is metabolised. Halothane is metabolised 20% compared to isoflurane which is 0.2% metabolised. Therefore, risk of reaction and chances of hepatocellular injury is more with halothane.

### III. Isoflurane

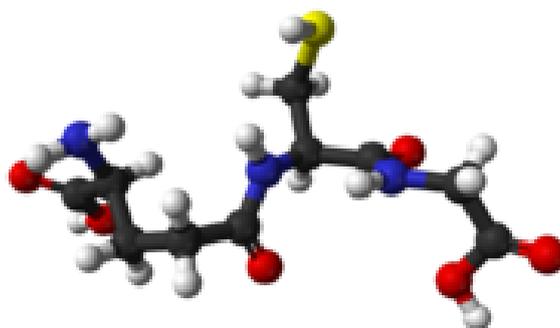


Isoflurane (2-chloro-2-(difluoromethoxy)-1,1,1-trifluoroethane) is a halogenated ether used for inhalational anaesthesia. Together with enflurane and halothane, it replaced the flammable ethers used in the pioneer days of surgery. Its use in human medicine is now starting to decline, being replaced with sevoflurane, desflurane and the intravenous anaesthetic propofol. Isoflurane is still frequently used for veterinary anaesthesia. Isoflurane is always administered in conjunction with air and/or pure oxygen. Often nitrous oxide is also used.



An initial step in detecting liver damage is a simple blood test to determine the presence of certain liver enzymes in the blood. Under normal circumstances, these enzymes reside within the cells of the liver. But when the liver is injured for any reason, these enzymes are spilled into the blood stream. Enzymes are proteins that are present throughout the body, each with a unique function. Enzymes help to speed up (catalyze) routine and necessary chemical reactions in the body.

Among the most sensitive and widely used of these liver enzymes are the aminotransferases<sup>12</sup>. They include aspartate aminotransferase (AST or SGOT) and alanine aminotransferase (ALT or SGPT). Aspartate aminotransferase [AST] is non specific found in liver, muscle, heart, alanine aminotransferase [ALT] are specific which is only find in liver. These enzymes are normally contained within liver cells. If the liver is injured, the liver cells spill the enzymes into blood, raising the enzyme levels in the blood and signaling the liver damage. Serum or plasma concentration of alanine amino transferase [ALT] and aspartate aminotransferase [AST] are commonly used to assess hepatic injury .however, because the aminotransferase are not specific to the liver and can be released under other condition, it is often difficult to ascribe small abnormalities in their plasma activities to hepatic damage in addition, the aminotransferase, which are found mainly in periportal hepatocytes, the cell most prone to damage by halothane, alcohol, toxin, hypoxia<sup>13</sup>.



Rohm<sup>7</sup> conducted a study to assess the influence of total intravenous anaesthesia (TIVA) with propofol vs. anaesthesia with desflurane by measuring  $\alpha$ -GST. Forty-two patients scheduled for elective prostatectomy were randomly allocated to receive desflurane, and thiopental (desflurane group) or propofol and remifentanyl (TIVA group).depth of anaesthesia was guided by bispectral index. Plasma concentrations of GST and aminotransferase were measured before induction (T0), at the end of surgery (T1), as well as 2 hour (T2) and 24 hours (T3) postoperatively. Hemodynamic parameter and bispectral index document. He concluded that use of Propofol as a part of TIVA regimen seems to have no influence on hepatocellular function during and after the surgery. In contrast patients receiving desflurane shows a transient slight but significant increase of  $\alpha$ -GST to above normal upper limit after anaesthesia, although this was without further clinical relevance.

Ray et al<sup>13</sup> studied the effects of spinal anaesthesia on GST concentrations measured by specific radioimmunoassay in 33 patients undergoing intermediate orthopedic, general or gynaecological surgery. GST concentrations were measured before anaesthesia and 3, 6 and 24 h after induction of anaesthesia. Hypotension (systolic blood pressure <70% of pre-induction value) was rapidly corrected with i.v. ephedrine and found that mean duration of surgery was 41 min (range 11-80). No increase in GST concentration was observed at any time, but at 24 h GST concentration was significantly reduced ( $P<0.05$ ). One patient in whom hypotension was not treated developed a greatly increased GST concentration at 3 h. He concluded that there is no association between spinal anaesthesia and disturbance of hepatocellular integrity when hypotension does not occur or is rapidly corrected.

Darling et al<sup>3</sup> studied the effect of halothane or isoflurane anaesthesia on hepatic function in 30 ASA I–III patients aged 18–70 years undergoing lumbar discectomy. Hepatic function was assessed before anaesthesia, at the end of surgery, and at 3, 6, 24 and 48 hours after surgery using routine enzyme tests of hepatic function and mitochondrial aspartate transaminase (mAST) activity. Although serum mAST activities increased after surgery in both groups of patients, these increases were statistically significantly greater in the group that received halothane. The groups were similar with regard to other tests of hepatic function. Calculation of the ratio of serum enzyme activities compared to baseline values suggested that mAST is a sensitive marker of anaesthetic-induced hepatic injury. Fifteen patients were anaesthetized with isoflurane and 15 received halothane. The groups were similar in age, weight, gender, alcohol intake, duration of hypotension, minimum arterial pressure and dose of anaesthesia. All patients had liver function test values of less than twice the upper limit of the reference range prior to anaesthesia. In both groups, serum concentration of bilirubin increased significantly above baseline at 24 and 48 hours after anaesthesia. Serum ALT activity decreased significantly ( $p<0.05$ ) below baseline throughout the study period in the halothane group. ALP and GT.

Hülya et al<sup>14</sup> studied Effects of halothane, enflurane, and isoflurane on plasma and erythrocyte antioxidant enzymes and trace elements. Alterations of the normal redox balance in mammals might be attributed to increases of plasma free-radical concentrations and/or a disruption of the protective mechanisms. These conditions lead to damage to cellular structure by the mechanism of lipoperoxidation, particularly in the liver, kidney, and central nervous system. In this study, the effect of general anesthesia on the oxidative metabolism of human plasma and erythrocytes was investigated. Forty-five patients undergoing anesthesia by using halothane, enflurane, or isoflurane were included in this study. Blood samples were taken preoperatively, the first hour, the first day, and the third day after the operation. Superoxide dismutase (SOD) and glutathione peroxidase (GSH-Px) enzyme activities and trace elements such as cofactor copper (Cu), Zinc (Zn) and Selenium (Se) levels were measured in plasma and red blood cells. This review showed that halothane and enflurane administration increased the plasma GSH-Px activity and reduced zinc levels. In addition, they lowered SOD and GSH-Px activities and trace element levels on erythrocytes. Isoflurane had no effect on plasma antioxidant enzymes, but, similar to the other, isoflurane decreased the plasma zinc levels, erythrocyte SOD and GSH-Px activities and trace element levels.

## References

- [1]. Denno R, Takabayashi A, Sugano M, Awane M, Jin MB, Morimoto T, Tanaka K, Yamaoka Y, Kobayashi N, Ozawa K. The ratio of reduced glutathione/oxidized glutathione is maintained in the liver during short-term hepatic hypoxia. *J Gastroenterol.* 1995 Jun; 30(3): 338-46.
- [2]. Murray JM, Rowlands BJ, Trinick TR. Indocyanine green clearance and hepatic function during and after prolonged anaesthesia: comparison of halothane with isoflurane. *British Journal of Anaesthesia* 1992;68:168-171.
- [3]. Darling J. R., Sharpe P. C., Stiby E. K., McAteer J. A. et al. Serum mitochondrial aspartate transaminase activity after isoflurane or halothane anaesthesia *British Journal of Anaesthesia*, 2000, Vol. 85, No. 2.
- [4]. Beckett, G.J. and Hayes, J.D.. Glutathione S-transferase measurements and liver disease in man. *J. Clin. Biochem. Nutr.*, 1987; 2, 1-24.
- [5]. Hussey AJ. Plasma glutathione s-transferase concentration as a measure of hepatocellular integrity following a single general anesthesia with halothane, enflurane or isoflurane. *Br J Anesth* 1988; 60, No.2, 130-135.
- [6]. Allan LG, Hussey AJ, Howie J, Beckett GJ, Smith. Hepatic glutathione S-transferase release after halothane anaesthesia: open randomized comparison with isoflurane. *Lancet.* 1987 Apr 4; 1(8536): 771-4.
- [7]. Röhm KD, Suttner SW, Boldt J, Schöllhorn TAH and Piper SN. Insignificant effect of desflurane–fentanyl–thiopental on hepatocellular integrity – a comparison with total intravenous anaesthesia using propofol–remifentanyl. *Eur. J. Anaesthesiology.* 2005, 22: 209-214.
- [8]. Russell A. Van Dyke, PhD. Hepatic Centrilobular Necrosis in Rats after Exposure to Halothane, Enflurane, or isoflurane. *Anesth Analg* 1982; 61:812-819
- [9]. Sherlock S. Results of a controlled clinical trial of glutathione in cases of hepatic cirrhosis. *Gut* 1965; 6: 472-476.
- [10]. Gelman S. Liver circulation and function during isoflurane and halothane anesthesia. *Anesthesiology* 1984; 61(6): 726-30.
- [11]. Hayes P C, Bouchier I A, and Beckett G J. Glutathione s-transferase in human health and disease. *Gut.* 1991 July; 32[7]: 813-818.
- [12]. Kidd PM. Glutathione: systemic protectant against oxidative and free radical damage. *Altern Med Rev* 1997; 1: 155-176.
- [13]. Ray DC, Robbins AG, Howie AF, Beckett GJ, Drummond GB. Effect of spinal anaesthesia on plasma concentrations of glutathione S-transferase. *Br J Anaesth.* 2002 Feb; 88(2):285-7.
- [14]. Hulaya Turkan, Neslihan Bukan, Ahmet Sayal, Ahmet Aydin and M Hakan Burkan. Effect of halothane, enflurane and isoflurane on plasma and erythrocyte antioxidant enzymes and trace elements. *Journal of Biological Trace Elements Research.* 2004; 102: 105-112.