

# Effect of infection with the giant liver fluke (*Fasciola gigantica*) on the Superoxide dismutase (SOD) in liver tissue naturally infected with parasite.

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## Abstract

The current study was conducted with the aim of assessing the level of antioxidant (SOD) in the tissues of worms, and tissues of liver cows infected with *Fasciola gigantica* and uninfected livers in order to determine the effects of *Fasciola gigantica* infection. Through the current study, which extends from 12/12/2021 to 30/7/2022, 1422 cows and sheep livers were examined from butcher shop in Baquba city, Diyala province. One hundred and seventy four of livers for cows, and 1248 livers of sheep were collected with 14 non-infected liver in current study. In this study, 25 liver samples were infected with *Fasciola gigantica* belong to cows with absence of infection in sheep. Pieces of infected and non infected livers (14 non infect liver) samples were taken and homogenized. The activity of the enzyme Superoxide dismutase (SOD) was measured by ELISA. The results of the current study showed that there is a clear significant difference in the concentration of this enzyme for each of worms and infected liver ( $P < 0.05$ ). The results also showed that there is a significant difference between the infected group and the control group in all the studied areas ( $P < 0.05$ ).

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## Keywords

Superoxide dismutase, *Fasciola gigantica*

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Fasciolosis which is a disease caused by a parasite known as *Fasciola hepatica* and *Fasciola gigantica*. It is a common parasitic disease in the tropics (Ibrahim, 2017). The life cycle of this genus includes final hosts includes cows, sheep goats and many other ruminants while the intermediate host is the snails (Kurnianto, 2022), so the disease is prevalent in areas where grazing, irrigation and water marshes are prevalent, and where the snails of the genus *Lymnaea* is most frequent (Dalton et al., 1999). The disease is characterized by being of a chronic, acute or acute-like type and affects the bile ducts of the liver and is accompanied by odema, edema, anaemia, anorexia and general asthenia leading to death (Itagaki et al., 2022). Therefore, this disease causes huge economic losses in some

countries that depend for their daily income on livestock and their products either by reducing their products and failing the quality of products or by the death of animals (Mpisana., 2022). The parasite may cause significant damage to the liver of the infected person and thus cause liver dysfunction (olivares\_ feeretti et al., 2022). In some cases, infection with this parasite may lead to secondary infection with bacteria such as bacillary icterohemoglobinuria. In livestock resulting from infection with types of bacteria in the bacteria *Clostridium* (Wagari, 2021).

Infection with this parasite leads to physiological, immune and biochemical changes of the host if the infection stimulates the host's immune system leading to

the creation of immune and inflammatory responses as well as biochemical mechanisms and infection with this parasite has sometimes been considered a factor leading to the occurrence of oxidative effort (Jabbar, 2022).

Infection with the *Fasciola* causes the release of active oxygen molecules (ROS) which cause damage to the tissue resulting in damage to the tissue at the site of infection and that the increase in the immune reaction in the tissue to the presence of the parasite by releasing these molecules leads to exposing the tissue to oxidative effort and the fibrosis resulting from the injury may be another source of oxidative effort through the development of tissue to produce Free radicals to face injury and this requires the fabric to protect itself from those damages and free radicals so it may try to produce antioxidants to neutralize the effect of these harmful agents (Mendes et al., 2013; Bahrami et al., 2014)

AL-Juboury (2008) recorded that 36.73% of the percentage of infection in cows in Karbala province. Mikaeel (2020) explained that the incidence of *Fasciola gigantica* in Duhok was as follows in goats 6.3 and in sheep 8.9% and in the serological examination of the samples tested. while in Sulaymaniyah it, Raouf et al (2020) using PCR and out of 100 samples liver crop from slaughterhouses and 100 samples of feces for sheep and cows and the incidence rate was 15%. Among Shahathe et al (2021) said that the local infection rate was 43.5% and the highest year of infection in Ramadi was 69.9%.

The body has many antioxidant enzymes such as SOD (Desai et al., 2010). The enzyme was classified as a mineral protein as it was first isolated in 1959 and described as a copper-containing protein and is one of the enzymes involved in the analysis of metabolic toxic products in the cell, it removes the free radical of O<sup>-</sup>. By speeding up the conversion rate of its conversion to H<sub>2</sub>O with the help of some metals such as selenium, copper and zinc. ( Fukai and Ushio-Fukai, 2011) , there are three isotopes in mammals that vary according to their cellular distribution and associated minerals, including Cu/Zn-SOD, which is mainly found in the Mn-SOD cytoplasm, while the Ec-SOD form is located outside the cell (Yen et al, 2009). The present study evaluate the level of SOD in worm and liver tissues

## Materials and Methods

### Livers collection

In this study, 1422 cows and sheep livers were examined from the butchers in the city of Baquba with the special information of each infected animal taken. The samples were collected for the period from 12 of December 2021 until 30 of July 2022. The samples were collected with 174 cows and 25 infected livers and 1284 sheep livers and no infection were appeared in. The samples were then transferred to the biology Laboratory at the College Education for Pure Sciences Department of biology by plastic bags and the autopsy process was carried out.

The infected organs were thoroughly washed with water for the purpose of getting rid of blood, impurities and substances attached to them resulting from the slaughter process and placing the liver in a sterile dish where the outer surface of the liver was sterilized and then the liver was dissected to investigate worms.

The worms were isolated from the infected livers. The samples were then preserved by placing them in a PBS regulated phosphate solution. Then it is frozen 20- until the homogeneous extract of worms is prepared.

Three tissue biopsies of liver (5 g per piece) were taken, (bile duct, closed to infected area, far from the infected area) were taken. A biopsy was also taken from the tissue of uninfected livers. The samples are placed in t containers with phosphate buffer (PBS) and placed at a temperature of -20 °C. The worms and livers were crushed and then 3ml of PBS was added and centrifuged at a rate of 3000 rpm for 30 minutes and at a temperature of 4 ° C. The floating part of the sample was taken. The concentration of SOD was measured and according to the manufacturer's instructions for the kit

## Results and discussion

### Antioxidant Concentration (SOD)

A clear significant difference in the concentration of this enzyme is observed between the studied liver areas, namely the bile duct region, the area near the site of injury and the area far from the injury ( $P < 0.05$ ). The results also showed that there is a significant difference between the infected group and the control group in all the studied areas ( $P < 0.05$ ).

Table 1: shows the concentrations of the SOD enzyme in worms and studied areas.

Sex	The rate of concentration in worms	The rate of concentration in the bile duct	Concentration rate in the area near the injury	Concentration rate in the area away from injury	P value
Male	0.241	0.361	0.641	0.415	0.048
Female	0.241	0.364	0.655	0.781	0.028
Total	0.241	0.362	0.653	0.598	
Male control			0.877		0.036
Female control			0.866		0.042
Total			0.872		
P value	0.006	0.022	0.028	0.026	

Furthermore, the results show the concentration of the enzyme superoxide dismutase SOD in the tissues of the livers of animals affected by *Fasciola gigantica* under study decreased compared to the control groups, where the results recorded the concentration of SOD in the livers of infected animals 0.362 µg/L, while in the control groups the results recorded the concentration of SOD in the livers of healthy animals 0.872 µg/L. What the results showed is that the concentration of SOD is high in the parasite 0.241 µg / l compared to other areas of the affected liver tissue, where the results recorded a concentration of SOD in the bile duct of 0.362 µg / l, the area near the site of infection 0.653 µg / l and the area far from the site of injury 0.598 µg / l as shown in the table (1). The decrease in the concentration of SOD may be As a result of an increased amount of reactive oxygen and hydroxyl roots, it was known that the chemical composition of SOD was disrupted and that the low concentration of SOD led to a rise in super-aniones and a decrease in major antioxidant activities (Nabi et al., 2017).

Brown et al. (1997) stated that SOD is found in almost all worms, and it also secretes it as metabolic products, thanks to the fact that the hosts in which these worms are present secrete this enzyme to neutralize defensive oxidants, hence we find that the current study recorded the presence of a level of SOD in the weaving of isolated worms. The results of the study were consistent with the study conducted by Nabi et al. (2017) with a lower proportion of SOD antioxidant levels in the intestines and liver of parasite-infected fish compared to uninfected fish. The current study Seraslan et al. (2005) also agreed on a decrease in the antioxidant SOD. In the group affected by cutaneous leishmaniasis compared with the control group. Among Biswas et al. (1997) there was a significant decrease in the value of SOD in patients with calazar. While the current study did not agree with the results of Miltra et al (2017) which indicated that SOD is rising sharply due to reactive oxygen types ROS It works to enhance virulence against liver openwork by protecting the parasite from oxidative stress. The results of the

current study also did not agree with the results of the study conducted by Rubio et al. (2019) for a significant rise in SOD levels in patients with skin leishmaniasis compared to control groups as this enzyme is the first line of defense against the root O2. which stimulates its disintegration into H2O2 and then into oxygen and water and these enzymatic systems work to remove toxins from the root hydrogen peroxide where SOD also works by removing ROS toxins to remove oxidative stresses. Other studies have also shown SOD It increases the relaxation effectiveness of the blood vessels that adding SOD to connective cells could be more resistant to leishmaniasis (Rubio et al., 2019). The present study concludes that the infection with giant liver fluke play a role in unbalance of antioxidant of host.

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