



Study the effect of fungal filtrate *Cladosporium sphaerospermum* on production of vital it in Chicks compounds and the possibility of biodegradation

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Abstract

This study was carried out in the veterinary hospital-Kufa in order to study the fungal isolates filtrate of *Cladosporium sphaerospermum* in Al-Najaf governorate on the production of vital compounds and their effect on chicks, and the effect of biodegradation by using the biological agents. The results of the filtrate analysis of the two isolates *C. sphaerospermum* showed a difference in the production of vital compounds. The fungal filtrate isolates of C1 were given the vital compounds H24O2, C13H24O2, C18H34O2, C14H30O3S, C12H24, C14H29I, C14H26O4, C10H21I, C13H28, C13H28, C10H21I, C13H28. While the fungal filtrate isolate of C2 was given the vital compounds H2O2, C17H32O2, C18H34O2, C11H21BrO2, C27H44O4. The results showed that the internal organs of broiler chicks were affected after 28 days of treatment with two isolates of C1 and C2 isolates. The results showed that the isolates filtrate had an effective effect on the internal organs of the broiler chicks after the filtrate of two isolates, after the oral administration of the isolates filtrate, where the treatment of isolates filtrate *C. sphaerospermum* significant effects in the weight of liver, kidney and heart compared to the treatment of control. The treatment of isolate filtrate of C1 gave the rate of increase in weight of liver, kidney and heart amounted 4.66 and 1.76 and 0.79 g respectively, while the treatment of isolate filtrate of C2 gave weights rates of 3.66 and 1.26 and 0.77 g respectively, compared to control treatment, which gave weights of 2.78 and 0.68 and 0.53 g, respectively. The treatment of fungal growth to fungal filtrate showed significant differences in the weight values of internal organs when compared to the treatment of the growth of fungal filtrate alone, which did not differ from control treatment. The treatment of growth *P. ostreatus* fungi showed a significant improvement in liver, kidney and heart weights amounted 3, 1.13 and 0.63 g, respectively. The treatment of growth *P. ostreatus* isolate filtrate fungi of C2 showed also a significant improvement in liver, kidney and heart weights. 3.01, 0.68 and 0.53 g, respectively.

The results showed that fungus filtrate had an effective role in the effect, and the effects of the treatment with fungal filtrate vary in the manner of oral administration in the effectiveness of enzymes. The treatment of fungal isolates filtrate *C. sphaerospermum* showed significant effects in GOT, GPT and ALP compared to the rest of the treatments. The treatment with the C1 fungus gave GOT, GPT, and ALP a mean of 60.31, 4.5 and 22.4 respectively, while the treatment of filtrate fungus C2 82.12, 4.9 and 26.89, respectively, compared with control treatment, which gave 102.6, 9.3 and 30.91, respectively.

Fungi play an important role in the biodegradation of fungal filtrate. The treatment of fungus growth to fungal filtrate showed a significant differences in enzyme activity when compared with the treatment of fungal filtrate alone, which did not differ from the control treatment. The treatment of the fungus growth of *P. ostreatus* fungi to the fungus filtrate of C1 led a significant improvement amounted 90.12, 6.2 and 28.23 respectively, while the treatment of growth of *P. ostreatus* fungi to the fungal filtrate of C2 showed a significant improvement amounted 99.11, 9 and 28.07 respectively.

The results showed that the fungal isolates filtrate of *C. sphaerospermum* has an effective role in influencing the blood picture parameters. The effect of the treatment with the fungal filtrate varies with the oral administration method on the blood picture parameters in the broiler chick compared to the rest of the treatments. The treatment of fungal filtrate of C1 gave a decrease in RBC, PCV and Hb amounted 2.1, 22.62 and 5.11 respectively, while the treatment of fungal filtrate of C2 gave values of 2.13, 27.33 and 5.96, respectively, compared to the control treatment, which gave values of 2.95, 38.55 and 10.1 respectively.

The fungal isolate filtrate of C1 caused an increase in WBC amounted 30.11 while the treatment of fungal isolate filtrate of C2 gave a value of 28.6 while the control treatment was given 23.13. The fungus also played an important role in the biodegradation of vital compounds of the fungal filtrate where the treatment of growth fungi showed a significant difference in the weight rates of internal organs when compared to the treatment of growth of fungal filtrate alone, which did not differ from the treatment of control. The treatment of growth of *P. ostreatus* fungi to fungal filtrate of C1 resulted in a significant improvement of 2.85, 35, 9 and 23.01 respectively. The treatment of growth of *P. ostreatus* fungi to the fungal filtrate of C2 showed significantly improved reached 2.3, 35.66, 7.09 and 23.33, respectively.

Keywords: *Cladosporium sphaerospermum*, chicks

Introduction

Cladosporium is an important fungus that has the ability to survive in the soil and its ease in air. *Cladosporium*

cladosporioides is common fungi in the air and present in all external and internal environments (Bensch *et al.*, 2010; Flannigan *et al.*, 2016)^[3, 7]. It is highly dispersed in the air

via fungal spores (Deshmukh and Rai, 2005)^[5]. Mushrooms infect humans and animals, causing diseased kidneys and liver (Pokrzywa *et al.*, 2007)^[14], allergies and skin diseases (Denis *et al.*, 2015)^[4] Huang *et al.*, 2016)^[8] and asthma (Baxi *et al.*, 2016)^[2], also Ng *et al.* (2012)^[12] reported that it affects humans and animals causing subcutaneous disease called phaeohyphomycosis.

Al-Matar and Essam (2016)^[1] reported that fungus produces compounds for pharmaceutical, industrial and agricultural use, The fungus of *C. cladosporioides* may produce the compounds of p-methylbenzoic acid, ergosterol peroxide and calphostin, and the enzymes of pectin methylesterase, polygalacturonase, and chlorpyrifos hydrolase. The use of microorganisms to biodegradation and absorb metabolic materials is one of the most efficient and safe methods in liquid media, several studies have indicated that they are added to the animals as biologic supports. Several organisms were used to destroy the AFB1 in liquid media, including yeasts, ointments, bacteria, actinomycosis and algae. Lee *et al.*, (2003)^[11] indicated the ability of bacteria *Lactobacillus rhamnosus* GG to remove and eliminate toxins in laboratory and field experiments and the importance of this fungus and the lack of studies that highlight the importance of this study, which aims to study the effect of fungal filtrate of *C. sphaerospermum* in broiler chicks.

Due to the importance of this fungus and the few studies that highlight the importance of this study, which aims to study the effect of fungal filtrate of *C. sphaerospermum* in broiler chicks.

Materials and Methods

C. sphaerospermum Isolates

Two isolates of *C. sphaerospermum* were isolated from wheat seeds and Air. The study of two isolates of *C. sphaerospermum* filtrate on the production of vital compounds and their effect in the rate of internal organs weights, Physiological and Biochemical of chicks blood.

The inoculums of *C. sphaerospermum* were in the form of disks, prepared using a sterile cork poorer (5 mm). The disks were obtained from homogenous growth of 5 days old cultures grown in Potato Dextrose Agar (PDA) medium at 28°C. All the three conducted experiments in this study were complete randomized design (C.R.D.) with three replicates. The experiments were carried out in the veterinary hospital-Kufa

Preparation of fungal filtrates

The culture medium of P.D.B. was prepared according to the method described by Dewan (1989)^[6], after filtrate using the filter paper 1.No Whatman several times after it was filtered with a precise filter diameter of 0.22 mM. The crude filtrate is full strength (100% concentration). Concentration was prepared 30% of the crude filtrate of the fungus by taking 3 ml of each leachate and adding it to 7 ml of culture medium.

Test the effect of fungus isolates of *C. sphaerospermum* that selection on the production of biochemical compounds and the possibility to biodegradation by the fungus of *A. niger* and *P. ostreatus*

The fungal filtrate was prepared at the age of 28 days and *A. niger* and the fungus of *P. ostreatus* was inoculate with a 0.5 cm disc for each flask and 50 mL container of both *C. sphaerospermum* isolates as well as fungal filtrate without

inoculation. After 28 days the fungal filtrate was analyzed with GCMASS device in the Ministry of Science and Technology/ Environment and Water Department.

The compounds were known and their proportions before and after the treatment of the two fungi.

Test the efficacy of fungal isolates filtrate of *C. sphaerospermum* that selected on the chicks of white chickens

A white Belgian chick (type of Rose) from Najaf hatchery (a 3-day-old) was prepared from a field of veterinary medicine and supervised by veterinarians from the Veterinary Hospital of Kufa University, which was placed in cages in groups and adapted to the temperature and the lighting was appropriate, the treatments were the oral administration of chicks of 1 mL of filtrate/ kg of bird weight (30% filtrate concentration), taking into consideration the work of comparison with the oral administration by distilled water. The treatments were as follows:

- Distilled water (Control),
- Fungal filtrate *C. sphaerospermum* (C1),
- Fungal filtrate *C. sphaerospermum* (C2),
- Fungal filtrate *C. sphaerospermum* (C1) + *A. niger* fungi,
- Fungal filtrate *C. sphaerospermum* (C2)+ *A. niger* fungi,
- Fungal filtrate *C. sphaerospermum* (C1)+ *P. ostreatus* fungi,
- Fungal filtrate *C. sphaerospermum* (C2)+ *P. ostreatus* fungi.

A total of 95 chicks were prepared, divided into seven groups of three replicates, each containing five animals. The dosage was four weeks at a dose per week. During this period, clinical symptoms were observed. (EDTA) and the other part was placed in tubes containing an anticoagulant (EDTA), to conduct the physiological and biochemical blood tests the chicks were then slaughtered and vivisectioned by opening the abdominal cavity, and then took parts of the organs of the liver and kidney and heart and kept in formalin with a concentration of 10%.

Parameters studied

A. Physiological blood Parameters

1. Calculation of total hemoglobin concentration (Hb total)

Place 5 ml of Darbkin solution into a clean test tube and add 0.02 ml of drawn blood. The tube was well fed and then left for 10 minutes, hemoglobin meter was then reset with distilled water plus Darbkin solution. The tube was then placed in the device and was read in gm for every 100 ml with a wavelength of 540 nm.

2. Calculate the white blood cell count (W.B.C)

Place 0.4 ml of the solution in a clean test tube and place 0.02 ml of blood drawn by a Sali pipette and mix well, after the first droplets were removed, drop it to the Haemecytometer, then place the slide cover on it and leave to settle for 2 minutes, And tested under 40 × magnification power. The number of white blood cells was then calculated in the large squares in the angles of the cell count according to the following equation: W.B.Cs/mm³= Number cells Counted x 50

3. Calculation of packed cell volume (P.C.V.)

A quantity of blood was collected in capillary tubes leaving 15 mm of each empty tube. One end was closed with artificial clay and tubes were placed in the micro centrifuge at 11000 cycles/ min for 5 minutes. I then read the percentage of the blood mass (PCV%) by the inserted ruler (Hematocrit reader).

B. Biochemical blood parameters

Determination of the efficacy of alkaline Phosphatase (ALP)

The efficacy of the enzyme was estimated using a standard sample. In each of the three test tubes, 2 mL of substrat buffer solution, consisting of carbonat-bicarbonate, dinatriumphhenyl phosphate and PH = 10, was evaluated. The tubes were left in the incubator for 5 minutes at a temperature of 37 °C. The first tube was added 50 microliters of serum and 50 µl of phenol was added to the first tube and incubated for 15 minutes at 37 °C. Add 0.5 ml of Sodiumarsenate, antipurine Amino-4 inhibitor solution to each tube and apply well for 0.5 min of Fibrigenase (Kalum ferricynide) solution, in addition to the third pipe (Blank) 50 µl distilled water. The tubes were well bounced and left for 15 minutes in a dark place. Determine the optical absorption of the samples along the 510 wavelength using the optical spectrometer and calculate the enzyme's efficacy using the following equation

$$\text{Enzyme efficiency (Enzyme Unit/ 100 ml)} = \frac{\text{Reading the model} - \text{reading the blank}}{\text{Standard Readability}} \times N$$

N* = 20 units / 100 ml.

Determination of the enzyme efficiency of Glutamic Oxaloacetic Transaminase (GOT)

The efficiency of the GOT enzyme was estimated by using a standard kit, two tubes were tested: first 0.1 ml of serum and 0.1 ml of distilled water, 0.5 mL of GOT-buffer solution, consisting of a-oxoglutarate-1-aspartate-phosphat buffer, for 30 minutes, add 0.5 mL of dinitrophenyl hydracine 2-4 solution to the tubes and leave at room temperature for 20 minutes, prepare Sodium hydroxide (2N) solution and add 5 ml to the tubes, and leave the tubes five minutes. The absorption of samples to light was estimated by spectrophotometer on wavelength 546 and the readings were taken on a standard curve to extract the enzyme's efficiency.

Determination of the enzyme efficiency of Glutamic Pyuric Transaminase (GPT)

The same steps have been conducted in the GOT, with the exception of L-alanine in the enzyme-structured solution rather than L-aspartate and the readings were dropped on a standard GPT curve to extract the enzyme's efficiency.

Statistical analysis

Data from the three experiments were analyzed and analysis of variance ANOVA was performed using GenStat 12th edition (<https://www.vsni.co.uk/>). Means were compared according to least significant difference (Duncan) at 99% confidence (P≤0.01).

Results and Discussion

Fungi diagnosed

The results of the sequence analysis of the nitrogen bases showed the PCR products of isolated fungi in this study and using the BLAST site that the isolates were attributed to the *C. sphaerospermum* (Table 1).

Table 1: shows the molecular diagnosed fungi and the isolation region origin.

NO.	Fungi isolate	Region	Isolation
1	<i>C. sphaerospermum</i>	AL-Najaf	Seeds
2	<i>C. sphaerospermum</i>	AL-Najaf	Air
3	<i>C. sphaerospermum</i>	AL-Najaf	Plant residues
4	<i>C. sphaerospermum</i>	AL-Najaf	Air
5	<i>C. sphaerospermum</i>	AL-Najaf	Waste
6	<i>C. sphaerospermum</i>	AL-Najaf	Soil

Detection of raw filtrate contents of isolates of C1 and C2 of *C. sphaerospermum*

The results of chemical analysis showed by Thin Layer Chromatography technique (TLC) that the isolates filtrate of C1 and C2 have the ability to produce biochemical compounds. The results showed differences between the two isolates in the production of the biochemical compounds: C16H32O2, C13H24O2, C18H34O2, C14H30O3S, C12H24, C14H29I, C14H26O4, C10H21I,

C13H28, C13H28, C10H21I, C13H28 that mentioned in the table (2), as for the isolates of the fungus C2 gave vital compounds: C16H32O2, C17H32O2, C18H34O2, C11H21BrO2, C27H44O4 that mentioned in the table (3).

The isolation of fungi varied in their production of compounds and isolation C1 was the most productive based on the intensity of glare under ultraviolet light. The isolates variation in the production of biochemical compounds is due to the genetic structure of isolation.

Table 2: The contents of the fungal filtrate of fungus isolate of C1

No.	Compound site	Vital compound	Similarity	Time appearing	Concentration	Volume
1	5	C16H32O2	87	19.706	46.57	37.09
2	6	C13H24O2	75	20.947	1.02	1.52
3	7	C18H34O2	92	21.477	36.35	31.20
4	8	C16H32O2	85	21.633	7.20	12.49
5	10	C14H30O3S	81	23.641	0.71	1.86
6	11	C12H24 C13H28 C10H21I	82	24.498	1.31	2.86

		C13H28 C14H26O4				
7	12	C14H29I C10H21I	78	25.441	1.83	3.11
8	13	C13H28	80	26.520	1.50	2.27

Table 3: The contents of the fungal filtrate of the fungi isolate of C2

No.	Compound site	Vital compound	Similarity	Time appearing	Concentration	Volume
1	3	C16H32O2	86	19.624	39.70	34.77
2	5	C17H32O2	77	20.940	2.11	4.09
3	6	C18H34O2 C16H30O2	87	21.383	17.80	16.70
4	7	C16H32O2	86	21.592	20.96	21.99
5	10	C11H21BrO2 C27H44O4	75	27.224	9.15	10.52

The biodegradation of fungal isolates filtrate of C1 and C2 of the fungi of *C. sphaerospermum* by *Aspergillus niger* and *Pleurotus ostreatus*

The treatment of growth of *A. niger* fungi to isolates filtrate of C1 and C2 of the of *C. sphaerospermum* fungi both independently led to the production of new vital compounds (Table 4 and 6), also the treatment of growth of *P. ostreatus* fungi to isolates filtrate of C1 and C2 of the fungi of *C. sphaerospermum*, led to the production of new vital compounds, this result was consistent with the results of (Lee *et al.*, (2003) [11].

Test the efficacy of the isolates filtrate of C1 and C2 of fungus of *C. sphaerospermum* treated with *A. niger* and *P. ostreatus* and not on broiler chicks

Effect of the isolates filtrate of C1 and C2 of fungus of *C. sphaerospermum* treated with the fungus of *A. niger* and *Pleurotus ostreatus* and not treated in the internal organs weight rate

Table 4: biodegradation of fungal isolate filtrate of C1 by the fungi of *A. niger*

No.	Vital compound	Concentration	Volume
1	C16H32O2	22.11	37.09
2	C18H34O2	19.74	31.20
3	C16H32O2	1.01	12.49

Table 5: biodegradation of fungal isolate filtrate of C1 by the fungi of *P. ostreatus*

No.	Vital compound	Concentration	Volume
1	C16H32O2	20.63	32.09
2	C18H34O2	15.45	28.11
3	C16H32O2	1.00	9.39

Table 6: biodegradation of fungal isolate filtrate of C2 by the fungi of *P. ostreatus*

No.	Vital compound	Concentration	Volume
1	C16H32O2	30.10	12.87
2	C18H34O2 C16H30O2	11.21	12.46
3	C16H32O2	18.32	14.16

Table 7: biodegradation of fungal isolate filtrate of C2 by the fungi of *A. niger*

No.	Vital compound	Concentration	Volume
1	C16H32O2	16.70	9.71
2	C18H34O2 C16H30O2	8.18	11.10
3	C16H32O2	16.46	9.19

Test the efficacy of the isolates filtrates of C1 and C2 of the fungus of *C. sphaerospermum* treated with the fungus of *Aspergillus niger* and *Pleurotus ostreatus* and not treated on broiler chicks

The results of the table (8) Indicated that the internal organs of broiler chicks were affected after 28 days of treatment with the isolates filtrate of C1 and C2 isolates. The results showed that the isolates filtrates had an effective role on the internal organs of the chick broilers after the oral administration of the two isolates filtrates, the treatment of fungal isolates filtrate of *C. sphaerospermum* significant effects in weight of liver, kidney and heart compared to the control treatment as given treatment isolate filtrate of C1 rate of increase in weight of the liver and kidney and heart amounted 4.66 and 1.76 and 0.79 g respectively, while the treatment of isolates filtrate of C2 gave the weights rates reached 3.66, 1.26 and 0.77 g respectively, compared to the control treatment, which gave weights rates of 2.78, 0.68 and 0.53 g respectively.

Table 8: Effect of the isolates filtrate of C1 and C2 of fungus of *C. sphaerospermum* treated and non-treated of fungus of *Aspergillus niger* and *Pleurotus ostreatus* in the rate of internal organs weights (liver, kidney, heart) of the chicks.

Treatments (fungal isolates)	Organs weights		
	liver	kidney	heart
<i>C. sphaerospermum</i> (C1)	4.66a	1.76a	0.79a
<i>C. sphaerospermum</i> (C1) + <i>A. niger</i>	3.01b	1.50b	0.66b
<i>C. sphaerospermum</i> (C1) + <i>P. ostreatus</i>	3.00b	1.13b	0.63b
<i>C. sphaerospermum</i> (C2)	3.66a	1.26b	0.77a
<i>C. sphaerospermum</i> (C2) + <i>A. niger</i>	3.33b	1.40b	0.61b
<i>C. sphaerospermum</i> (C2) + <i>P. ostreatus</i>	3.01b	0.91c	0.56c
Control	2.78c	0.68c	0.53c

a, b Means with the different superscripts along the row are significantly different (P≤0.01)

The results in the table showed that the fungus of *Aspergillus niger* and *Pleurotus ostreatus* had an important role in biodegradation the vital compounds of the isolates filtrates of C1 and C2.

The treatment of the fungus growth to fungal filtrate showed significant differences in the weight values of internal organs when compared to the treatment of the of the fungal filtrate growth alone, which was not different from the treatment of control, the treatment of the growth of *A. niger* fungi to the isolate filtrate of C1 reduced the effect of fungal filtrate significantly in the weight of liver, kidney and heart reached 3.01, 1.5 and 0.66 g, respectively, while the treatment of the growth of *P. ostreatus* fungi to isolate filtrate of C1 showed a significant improvement in liver, kidney and heart weights, reached 3, 1.13 and 0.63 g respectively.

The treatment of the growth of *A. niger* fungi to the isolate filtrate of C2 reduced the effect of fungal filtrate significantly in the weight of liver, kidney and heart reached 3.33, 1.4 and 0.61 g, respectively, while the treatment of the growth of *P. ostreatus* fungi to isolate filtrate of C2 showed a significant improvement in liver, kidney and heart weights, reached 3.01, 0.68 and 0.53 g respectively.

Table 9: the effect of the isolates filtrate of C1 and C2 of fungus of *C. sphaerospermum* treated and non-treated of fungus of *Aspergillus niger* and *Pleurotus ostreatus* in the enzymes efficiency rate.

Treatments (fungal isolates)	Enzymes		
	GOT	GPT	ALP
<i>C. sphaerospermum</i> (C1)	060.31c	4.5c	22.40c
<i>C. sphaerospermum</i> (C1) + <i>A. niger</i>	082.11b	5.2b	26.11c
<i>C. sphaerospermum</i> (C1) + <i>P. ostreatus</i>	090.12b	6.2b	28.23b
<i>C. sphaerospermum</i> (C2)	082.12c	4.9c	26.89c
<i>C. sphaerospermum</i> (C2) + <i>A. niger</i>	098.03b	9.1a	28.11b
<i>C. sphaerospermum</i> (C2) + <i>P. ostreatus</i>	99.11b ⁰	9.0a	28.07b
Control	102.60a	9.3a	30.91a

a, b Means with the different superscripts along the row are significantly different ($P \leq 0.01$)

The results of table (9) showed that fungus had an important role in the biodegradation of fungal filtrates. The treatment of fungi growth to fungal filtrate showed significant differences in enzyme efficiency when compared with the treatment of the growth of fungal filtrate alone, which did not differ from control treatment. The fungi growth treatment of *A. niger* to fungal filtrate of C1 significantly reduced the effect of fungal filtrate at 82.11, 5.2 and 26.11, respectively.

The fungi growth treatment of *P. ostreatus* to fungal filtrate of C1 showed a significant improvement of 90.12, 6.2 and 28.23 respectively. Also the fungi growth treatment of *A. niger* to fungal filtrate of C2 significantly reduced the effect of fungal filtrate amounted to 98.03 and 9.1 respectively, and 28.11, while the fungi growth treatment of *P. ostreatus* to fungal filtrate of C2 showed a significant improvement amounted to 99.11, 9 and 28.07 respectively.

The effects of fungal filtrate may be due to its containment of enzymes, toxins, antibiotics life and metabolic compounds affecting other experimental animals. The low concentration of enzymes is due to damage to the target organ and therefore to the effect of the necessary enzymes function.

The effect of the isolates filtrate of C1 and C2 of fungus of *C. sphaerospermum* treated and non-treated of fungus of *Aspergillus niger* and *Pleurotus ostreatus* in blood picture parameters

The results of the table (10) showed the effect of the isolates

The effects of fungal filtrate may be due to its containment of enzymes, toxins, antibiotics life and other metabolic compounds affecting experimental animals. The filtrates caused the stop of the necessary enzymes function and then accumulate a lot of these substances in them as toxins causing an increase in the weight of internal organs. This result is consistent with similar studies by Kubena *et al.*, 1990^[10].

Effect of the isolates filtrate of C1 and C2 of fungus of *C. sphaerospermum* treated and non-treated of fungus of *Aspergillus niger* and *Pleurotus ostreatus* in the enzymes efficiency rate.

Table (9) shows the effect of the isolates filtrate of C1 and C2 of fungus of *C. sphaerospermum* and the effect of the biodegradation of fungi in the enzymes efficiency rate after 28 days of treatment, where the results showed that the fungus filtrates play an effective role in the enzymes efficiency rate. The effect of the treatment on the fungal filtrate varied with the oral administration method in the enzymes efficiency. The treatment of the two isolates filtrate of *C. sphaerospermum* had significant effects in GOT, GPT and ALP compared with the rest of the treatments.

filtrate of C1 and C2 of fungus of *C. sphaerospermum* and the effect of the biodegradation of fungi in blood picture parameters of the chicks broiler after 28 days of treatment, where the results showed that the fungus filtrates of *C. sphaerospermum* play an effective role in the effect in blood picture parameters. The effect of the treatment on the fungal filtrate varied with the oral administration method in blood picture standards. The treatment of isolates filtrate of *C. sphaerospermum* had significant effects in blood picture parameters compared with the others treatments. The treatment of the fungus filtrate of C1 gave a reduced of RBC, PCV and Hb amounted 2.1, 22.62 and 5.11 respectively, while the C2 fungus filtrate treatment gave values of 2.13, 27.33 and 5.96, respectively, compared with the control treatment which gave values reached 2.95, 38.55 and 10.1 respectively, and the fungus isolate of C1 caused an increase of WBC reached 30.11, while the treatment of the fungus isolate of C2 gave a value of 28.6, while the control treatment gave 23.13.

The table also shows that fungi has an important role in biodegradation of vital compounds of fungal filtrate, the treatment of the fungi growth to fungal filtrates showed significant differences in the weight values of the internal organs when compared to the treatment of the fungal filtrate growth alone, which did not differ from the control treatment. The treatment of the growth of *A. niger* fungi to the fungal filtrate C1 led to reduce the effect of fungus filtrate significantly reached 2.73, 34.63, 6.1 and 23.47 respectively for RBC, PCV, Hb and WBC, respectively.

The treatment of growth of *P. ostreatus* fungi to fungal filtrate of C1 was showed significantly improved reached 2.85, 35, 9 and 23.01 respectively. Also the treatment of the growth of *A. niger* fungi to fungal

filtrate of C2 was significantly reduced to 2.26, 35, 6.81 and 24.1 respectively, while the treatment of growth of *P. ostreatus* fungi showed a significant improvement of 2.3, 35.66, 7.09 and 23.33 respectively.

Table 10: Effect of the isolates filtrates of C1 and C2 of *C. sphaerospermum* fungi treated and no treated with *Aspergillus niger* and *Pleurotus ostreatus*, in the blood picture parameters.

Treatments (fungal isolates)	blood picture parameters			
	RBC	WBC	PCV	Hb
<i>C. sphaerospermum</i> (C1)	2.10c	30.11a	22.62c	5.11c θ
<i>C. sphaerospermum</i> (C1) + <i>A. niger</i>	2.73b	23.47b	34.63b	6.10b θ
<i>C. sphaerospermum</i> (C1) + <i>P. ostreatus</i>	2.85b	23.01c	35.00b	9.00b θ
<i>C. sphaerospermum</i> (C2)	2.13c	28.60a	27.33c	5.96c θ
<i>C. sphaerospermum</i> (C2) + <i>A. niger</i>	2.26b	24.10b	35.00b	6.81b θ
<i>C. sphaerospermum</i> (C2) + <i>P. ostreatus</i>	2.30b	23.33c	35.66b	7.09b θ
Control	2.95a	23.13c	38.55a	10.10a

a, b Means with the different superscripts along the row are significantly different ($P \leq 0.01$)

The effects of fungal filtrate may be due to its containment of enzymes, toxins, antibiotics life and other metabolic compounds that affect experimental animals. The filtrates caused the stop of the necessary enzymes function and then accumulate a lot of these substances in them as toxins causing an increase in the weight of internal organs.

As for the decrease in the values of blood parameters, it has been explained that it is due to the effect of vital compounds such as toxins in the intestinal ability to absorb the iron element or toxic effect in the bone marrow, resulting in a decrease in hemoglobin and then reduce the formation of red blood cells, and accordingly, hemoglobin rate have decreased, and there is a positive correlation between the number of red blood cells and the value of the blood mass, reduced number of red blood cells is reflected in the values of the blood mass, which is lower than normal levels.

The reason of the increase in the number of white blood cells is to stimulate the immune system of animals. This result is consistent with the results obtained by Kubena *et al.*, 1997^[9].

Conclusion

The fungal isolates filtrate of *Cladosporium sphaerospermum* were production of vital compounds and their effect on chicks and *P. ostreatus* fungi play an important role in the biodegradation of fungal filtrate

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