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## Improved Antioxidant Capacity and Immune Function of Broiler Chickens Fed with Selenium-enriched *Candida utilis*

### ABSTRACT

The aim of this study was to assess the effect of selenium-enriched *Candida utilis* with high contents of organic selenium (Se) and glutathione (GSH) on growth performance, antioxidant capacity and immune function of broiler chickens. A total of 100 healthy 7-day-old male broiler chickens were randomly divided into 5 groups, and fed diets supplemented with (a) Na<sub>2</sub>SeO<sub>3</sub>, (b) *C. utilis*, (c) Se-enriched *Saccharomyces cerevisiae*, (d) Se-enriched *C. utilis*, and (e) the control without any supplements. The experiment lasted for 6 weeks and parameters were recorded on day 42. No significant differences in average daily gain were found among the 5 groups. However, Se-enriched *C. utilis* supplemented in the diet increased activities of glutathione peroxidase in the whole blood ( $p < 0.01$ ), catalase in the serum ( $p < 0.01$ ) and breast meat ( $p < 0.01$ ), and superoxide dismutase in the breast meat ( $p < 0.01$ ), as well as decreased contents of malondialdehyde in the serum ( $p < 0.01$ ), liver ( $p < 0.01$ ) and breast meat ( $p < 0.05$ ). Also, Se-enriched *C. utilis* improved titers of IgG ( $p < 0.01$ ), IgM ( $p < 0.01$ ), and IgA ( $p < 0.01$ ) in the serum, as compared to the control. All these results indicated that Se-enriched *C. utilis* was a good candidate of dietary supplement to improve the antioxidant capacity and immune function of broiler chickens.

### INTRODUCTION

Selenium (Se) is an essential nutritional trace element for human beings and animals owing to its critical role in cell metabolism (Schrauzer, 2006; Kieliszek & Błażej, 2013). Se is now being widely studied because of its antioxidant and immunity-associated capacities in many higher eukaryotic organisms (Burk, 2002; Tapiero *et al.*, 2003; Rayman, 2005; Hoffmann & Berry, 2008). In broiler chickens, Se is essential for the normal function of antioxidant and immune systems, and Se deficiencies are associated with diarrhea, pancreas atrophy, reproductive dysfunction, and immune or nerve damage (Yang *et al.*, 2016). Thus, it is necessary to ensure sufficient Se in the diet by adding Se supplements to reduce the incidences of these symptoms.

In June 2000, Se-enriched yeast was approved by the US Food and Drug Administration as a safe source of feed-supplemented organic Se for animals (Food and Drug Administration, 2000). Since then, Se-enriched yeast (also known as selenized yeast) has commonly been used as a feed additive in the raising of livestock and poultry (Surai & Fisinin, 2014; Baltić *et al.*, 2015; Shi *et al.*, 2017). *Saccharomyces cerevisiae* and *Candida utilis* are usually used for the preparation of Se-enriched yeast because they are capable of producing biomasses with high protein contents and can transform inorganic Se (toxic and low bioavailability) into organic Se (safer and bioactive) (Yin *et al.*, 2010; Wang *et al.*, 2012). In recent years, Se-enriched *C. utilis*



has attracted more and more attention owing to its higher intracellular glutathione (GSH) content and higher bioconversion rate from inorganic Se to organic Se than Se-enriched *S. cerevisiae* (Yang *et al.*, 2013; Kieliszek *et al.*, 2017).

According to the literature, Se-enriched *S. cerevisiae* has been shown to enhance the growth performance, antioxidant capacity, and meat quality of broiler chickens (Wang & Xu, 2008; Li *et al.*, 2017). Moreover, organic Se from selenized *S. cerevisiae* also favored to improve the immune status of broiler chickens, and a higher titer of IgM was obtained after Se supplementation (Boostani *et al.*, 2015). However, little is known about the effects of Se-enriched *C. utilis* on the raising of broiler chickens. In this study, we aimed to evaluate the effects of Se-enriched *C. utilis* on growth performance, antioxidant capacity, and immune function of broiler chickens. Additionally, the reason why Se-enriched *C. utilis* was more effective than other supplements in the diet was also elucidated.

## MATERIALS AND METHODS

### Selenium sources

Sodium selenite ( $\text{Na}_2\text{SeO}_3$ ) was purchased from Sinopharm Chemical Reagent Co., Ltd. (Shanghai, China). The commercial Se-enriched *S. cerevisiae* (2000  $\mu\text{g/g}$  Se and 5.0 mg/g GSH) was purchased from AngelYeast Co., Ltd. (Yichang, China). The cells of *C. utilis* (14.6 mg/g GSH) and Se-enriched *C. utilis* (1884  $\mu\text{g/g}$  Se and 13.1 mg/g GSH) were prepared using *C. utilis* SZU 07-01 according to the methods described in our previous studies (Wang *et al.*, 2012; Nie *et al.*, 2010). All the yeast cells supplied to the diet were in live conditions.

### Experimental design and chicken trials

In total, 100 healthy 7-day-old male white broiler chickens, with a mean initial body weight of  $108 \pm 8$  g, were purchased from a local farm in Suzhou, China. The chickens were randomly divided into 5 groups with 20 animals per group. The dietary supplements were designed as follows: group A, inorganic Se of  $\text{Na}_2\text{SeO}_3$ ; group B, *C. utilis* cells; group C, commercial Se-enriched *S. cerevisiae*; group D, Se-enriched *C. utilis*; and group E, the control without any supplements (Table 1). The nutrient contents of the basal chicken diet are shown in Table 2. The dose of Se in the Se-supplemented diet was maintained at 3.0 mg/kg, and the dose of *C. utilis* provided to group B was equivalent to that of Se-enriched *C. utilis* provided to group D.

**Table 1** – Supplements in the diet of broiler chickens.

Description of groups	Se <sup>a</sup>	GSH
Group A: $\text{Na}_2\text{SeO}_3$	+ (inorganic Se)	–
Group B: <i>C. utilis</i>	–	+
Group C: Se-enriched <i>S. cerevisiae</i>	+ (organic Se)	+
Group D: Se-enriched <i>C. utilis</i>	+ (organic Se)	+
Group E: control	–	–

+: Diet containing this composition; –: none of this component in the diet.

<sup>a</sup> The dose of Se in the diets was maintained at 3.0 mg/kg.

The broiler chickens were fed for 6 weeks in stainless steel cages in an air-conditioned animal facility with a constant temperature of  $27 \pm 1^\circ\text{C}$ , humidity of  $50 \pm 5\%$ , and a 12-h light/dark cycle. The chickens were allowed free access to feed and fresh water. The body weight of the broiler chickens was determined weekly throughout the experiment, and average daily gain was calculated for each chicken. All animal procedures were performed in compliance with protocols approved by the Committee for the Care and Utilization of Experimental Animals of Soochow University.

**Table 2** – Nutrient content of the basal diet \*.

Chemical composition	Content (g/kg)
Water	101.3
Crude protein	200.2
Crude fat	40.4
Crude fiber	49.8
Ash	80.7
Calcium	10.3
Available phosphorus	6.2
Lysine	13.2
Methionine and cystine	7.8

\*Supplied per kilogram of diet: vitamin A, 10 000 IU; vitamin D<sub>3</sub>, 4000 IU; vitamin K, 3 mg; vitamin E, 100 IU; vitamin B<sub>1</sub>, 3.5 mg; vitamin B<sub>2</sub>, 10 mg; vitamin B<sub>6</sub>, 4 mg; vitamin B<sub>12</sub>, 1 mg; nicotinic acid, 50 mg; pantothenic acid, 20 mg; folic acid, 2 mg; biotin, 0.2 mg; choline, 1000 mg; manganese, 80 mg; manganese, 100 mg; iron, 80 mg; copper, 8 mg; zinc, 80 mg; and iodine, 0.35 mg.

### Sample collection and preparation

At the end of the animal experiment (on day 42), the broiler chickens were sacrificed after being electrically stunned. Blood samples were collected through veins under the wings, and then divided into two portions. One portion was collected into a 10-mL heparinized tube and centrifuged at 3,000 *g* and 4°C for 15 min to obtain serum, and another portion was collected as the whole blood, which was used to determine the contents of Se and GSH, and glutathione peroxidase (GSH-Px) activity. Both blood samples were gently ejected into 5-mL Eppendorf tubes at  $-70^\circ\text{C}$ . The breast meat samples of each carcass were individually sliced into different sections. One section was kept at



4°C for the measurement of the drip-loss rate, and the other sections were vacuum-packed in plastic bags and frozen at -70°C for further analysis. The liver, thymus, spleen, and fabricius tissues were rapidly excised and rinsed with ice-cold isotonic saline, and then stored at -70°C before determinations.

The contents of Se and GSH in the whole blood, breast meat, and liver samples were determined by the methods described in the previous study (Wang *et al.*, 2012).

### Determination of the drip-loss rate of breast meat

For the quantification of the drip-loss rate of breast meat, the fresh samples (~10 g) cut from the carcass were immediately hung up in an inflated plastic bag. After storage at 4°C for 48 h, the samples were weighed again. The drip-loss rate was expressed as a percentage of the reduced weight to the initial weight (Honikel, 1998).

### Antioxidant capacity assay

Chicken breast meat or liver samples (1 g) were homogenized in 9 mL of ice-cold buffer solution (50 mmol/L Tris and 0.5 mmol/L EDTA, pH 8.0) using an Ultra-Turrax homogenizer (Tekmar Co. Ltd., Cincinnati, OH, USA) at 8,000 rpm for 10 s and centrifuged at 3,000 *g* and 4°C for 15 min. The supernatant was used to analyze the antioxidant capacity. The GSH-Px activity in the whole blood, catalase (CAT) and superoxide dismutase (SOD) activities, as well as the malondialdehyde (MDA) levels in serum, breast meat, and liver samples were assayed using appropriate commercial assay kits purchased from Nanjing Jiancheng Bioengineering Institute (Nanjing, China) in compliance with the manufacturers' instructions. The assays were conducted using spectrophotometric methods (UV2550, Shimadzu, Tokyo, Japan).

### Immunity assessment

The immune organs, thymus, spleen and fabricius, of each chicken were weighed. The immune organ indices were expressed as the ratio of the weight of the immune organ (thymus, spleen, or fabricius) relative to the chicken's body weight. The titers of immunoglobulins, such as IgG, IgM, and IgA, in the serum were determined by the double-antibody sandwich enzyme-linked immunosorbent assay method using commercial kits purchased from Suzhou Kechuang Biotechnology Co., Ltd. (Suzhou, China) in compliance with the manufacturers' instructions.

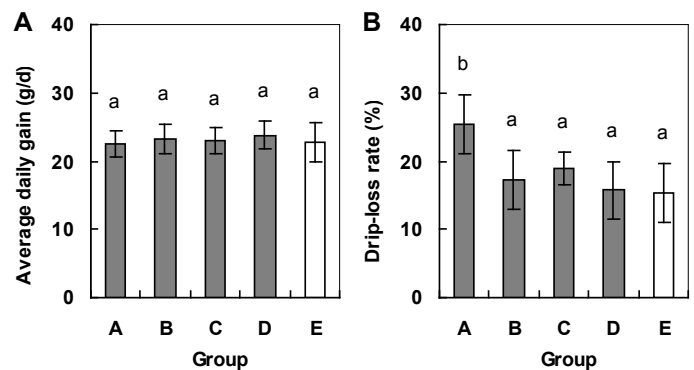
### Statistical analyses

All values were expressed as means  $\pm$  standard deviations. The Student's *t* test was employed to evaluate statistical significance, and samples with  $p \leq 0.05$  were considered to be statistically significant. Statistical calculations were performed using SPSS 17.0 software (SPSS Inc, Chicago, IL, USA).

## RESULTS

### Growth performance

The growth performance of broiler chickens fed with diets containing different supplements was evaluated by comparing the average daily gain of chickens. As shown in Fig. 1A, no significant differences in average daily gain were found among the five groups. That is, the growth of chickens fed with different supplements showed similar results relative to the control.



**Figure 1** – The growth performance (A) and drip-loss rate of the breast meat (B) of broiler chickens fed with different dietary supplements of  $\text{Na}_2\text{SeO}_3$  (group A), *C. utilis* (group B), Se-enriched *S. cerevisiae* (group C), and Se-enriched *C. utilis* (group D). The control (group E) represents no addition of supplement in the basal diet of broiler chickens. Bars labeled with different letters indicate significant differences ( $p < 0.05$ ), while the same letters indicate no significant differences ( $p > 0.05$ ), as compared to the control.

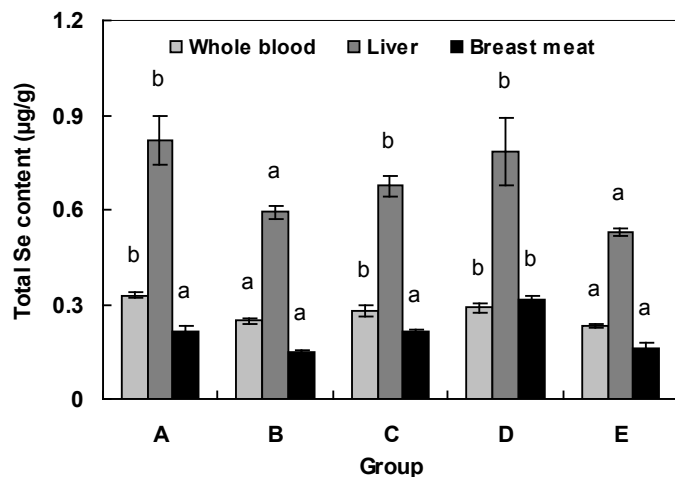
The drip-loss rate of chicken breast meat was also determined and compared. As shown in Fig. 1B, group A ( $\text{Na}_2\text{SeO}_3$ ) showed higher drip-loss rate ( $p < 0.05$ ) than that of the control, indicating that the water-holding capacity of breast meat decreased greatly in broiler chickens fed with inorganic Se. Chickens fed diets supplemented with yeast cells (whether selenized or not) showed no significant differences in drip-loss rate of the breast meat relative to the control.

### Se distribution

Total Se contents in the whole blood, liver, and breast meat of broiler chickens were determined. As shown in Fig. 2, no changes in Se contents in whole blood, liver and meat were observed in group B (*C. utilis* cells), when compared to the control. However, the diets supplemented with either organic Se



(group C and D) or inorganic Se (group A, Na<sub>2</sub>SeO<sub>3</sub>) significantly increased Se contents ( $p < 0.05$ ) in both whole blood and liver. Moreover, the Se content of the breast meat in group D (selenized *C. utilis*) increased by 95%, as compared to the control. According to the above results, only Se-enriched *C. utilis* simultaneously improved the Se contents in whole blood ( $p < 0.05$ ), liver ( $p < 0.05$ ), and breast meat ( $p < 0.05$ ) of broiler chickens.



**Figure 2** – Total Se content in the whole blood, liver, and breast meat of broiler chickens fed with different dietary supplements of Na<sub>2</sub>SeO<sub>3</sub> (group A), *C. utilis* (group B), Se-enriched *S. cerevisiae* (group C), and Se-enriched *C. utilis* (group D). The control (group E) represents no addition of supplement in the basal diet of broiler chickens. In the same tissue, bars labeled with different letters indicate significant differences ( $p < 0.05$ ), while the same letters indicate no significant differences ( $p > 0.05$ ), as compared to the control.

### Antioxidant enzyme activities

The effects of different dietary supplements on the activities of antioxidant enzymes, such as GSH-Px, CAT, and SOD, in blood and tissues of broiler chickens were investigated. As shown in Table 3, no obvious differences were found in the activity of GSH-Px in the whole blood between groups A (Na<sub>2</sub>SeO<sub>3</sub>), B (*C. utilis*), C (selenized *S. cerevisiae*), and the control. However, group D supplemented with Se-enriched *C. utilis* resulted in much higher GSH-Px activity in the whole blood, which increased by 41.7% ( $p < 0.01$ ) as compared to the control.

Moreover, supplements of Se and/or yeast in the diet, all increased the activity of CAT in the serum ( $p < 0.01$ ), while had no significant effects on CAT activity in the liver. As to the CAT in the breast meat, only Se-enriched *C. utilis* resulted in an increase of 72.4% ( $p < 0.01$ ) in CAT activity relative to the control, other supplements showed no significant differences in CAT activity. In addition, the activities of SOD in the serum and liver of broiler chickens showed no significant differences between the control and groups fed with diverse supplements. However, the activities of SOD in the breast meat increased when adding selenized *S. cerevisiae* or selenized *C. utilis* to the diet, which increased by 23.9% ( $p < 0.05$ ) and 43.6% ( $p < 0.01$ ), respectively, as compared to the control.

**Table 3** – Antioxidant capacities of broiler chickens fed with different dietary supplements.

Group	A	B	C	D	E
Supplements	Na <sub>2</sub> SeO <sub>3</sub>	<i>C. utilis</i>	Se-enriched <i>S. cerevisiae</i>	Se-enriched <i>C. utilis</i>	Control
<b>Whole blood</b>					
GSH-Px (U/mL)	2672±107a	2752±378a	2512±398a	3264±164B	2304±454aA
GSH (mg/L)	36.08±2.07a	45.57±2.93b	37.38±1.40a	47.57±3.35b	39.49±3.94a
<b>Serum</b>					
CAT (U/mL)	0.70±0.04B	0.77±0.07B	0.67±0.07B	0.83±0.08B	0.51±0.04A
SOD (U/mL)	1.43±0.02a	1.42±0.05a	1.42±0.11a	1.48±0.05a	1.26±0.24a
MDA (nmol/mL)	2.34±0.21b	2.04±0.19B	2.47±0.19b	1.49±0.15B	2.77±0.21aA
<b>Liver</b>					
CAT (U/g)	13.02±2.49a	13.76±1.62a	12.96±2.31a	14.62±2.16a	12.61±1.62a
SOD (U/g)	1.64±0.30a	1.75±0.33a	1.64±0.27a	1.90±0.29a	1.59±0.30a
MDA (nmol/g)	0.39±0.03a	0.34±0.03b	0.36±0.02b	0.29±0.03B	0.42±0.05aA
GSH (mg/g)	4.55±0.98a	5.25±0.78b	4.25±0.09a	7.19±0.92B	4.22±0.55aA
<b>Breast meat</b>					
CAT (U/g)	0.23±0.09a	0.27±0.09a	0.24±0.06a	0.30±0.01B	0.18±0.08aA
SOD (U/g)	0.51±0.05a	0.52±0.08a	0.53±0.03b	0.61±0.07B	0.42±0.08aA
MDA (nmol/g)	0.04±0.01a	0.03±0.01b	0.05±0.01a	0.03±0.01b	0.05±0.01a
GSH (mg/g)	0.18±0.02B	0.45±0.04B	0.09±0.01B	0.49±0.02B	0.07±0.01A

<sup>a,b,A,B</sup> In the same row, values with different lower-case letters indicate significant differences ( $p < 0.05$ ), with different capital letters indicate extremely significant differences ( $p < 0.01$ ), while the same letters indicate no significant differences ( $p > 0.05$ ).

GSH-Px, glutathione peroxidase; GSH, glutathione; CAT, catalase; SOD, superoxide dismutase; MDA, malondialdehyde.





### GSH and MDA contents

The contents of GSH in the whole blood, liver, and breast meat of broiler chickens fed with different dietary supplements were determined. As shown in Table 3, groups A ( $\text{Na}_2\text{SeO}_3$ ) and C (selenized *S. cerevisiae*) showed no significant differences in GSH contents in the whole blood and liver, as compared to the control (group E). However, supplements of *C. utilis* (group B) and selenized *C. utilis* (group D) both resulted in increases in GSH contents of the whole blood and tissues. The GSH contents increased by 15.4% ( $p<0.05$ ) and 20.5% ( $p<0.05$ ) in the whole blood, 24.3% ( $p<0.05$ ) and 70.3% ( $p<0.01$ ) in the liver, and 594% ( $p<0.01$ ) and 658% ( $p<0.01$ ) in the breast meat, respectively, as compared to the control. Thus, the supplementation of *C. utilis* and Se-enriched *C. utilis* to the diets contributed to the accumulation of GSH in blood and tissues of broiler chickens.

The intracellular MDA contents in serum, liver, and breast meat of broiler chickens were also assayed. As shown in Table 3, MDA in the serum significantly decreased by adding all the supplements ( $p<0.05$  for groups A and C;  $p<0.01$  for groups B and D) tested in this study, while no differences in contents of MDA in the liver were found when  $\text{Na}_2\text{SeO}_3$  was supplemented to the diet. In addition, MDA in the breast meat only decreased in the presence of *C. utilis* ( $p<0.05$ ) and Se-enriched *C. utilis* ( $p<0.05$ ), as compared to the control. According to the results shown in Table 3, it should be noted that the greater the GSH contents obtained with supplements, the lower the MDA contents were found in serum, liver, and breast meat of chickens. The Se-enriched *C. utilis* resulted in the lowest MDA contents in serum and tissues, indicating that broiler chickens fed with this kind of selenized yeast had much greater antioxidant capacities than those did not receive any dietary supplements.

### Immune functions

The immune functions of broilers, including the immune organ (thymus, spleen, and fabricius) indices and the titers of immunoglobulins (IgG, IgM, and IgA) in serum were determined and compared in Table 4. When the broilers were fed with Se-enriched *S. cerevisiae* as the dietary supplement, the indices of thymus ( $p<0.05$ ) and fabricius ( $p<0.01$ ) decreased significantly, while the spleen index increased ( $p<0.01$ ), as compared to the control. Besides, other dietary supplements ( $\text{Na}_2\text{SeO}_3$ , *C. utilis*, and Se-enriched *C. utilis*) showed no differences in the indices of immune organ of broiler chickens.

Moreover, the titers of IgG in the sera increased ( $p<0.01$ ) when the chickens were fed with Se-enriched *C. utilis*, other supplements showed no significant differences in IgG as compared to the control. The IgM titers both increased with the addition of *C. utilis* ( $p<0.01$ ) and Se-enriched *C. utilis* ( $p<0.01$ ) but decreased with selenized *S. cerevisiae* ( $p<0.01$ ). Different from titers of the above two immunoglobulins, the IgA titer increased significantly ( $p<0.01$ ) with all the supplements in the diet. Taken together, the supplementation of Se-enriched *C. utilis* in the diet resulted in the highest titers of IgG, IgM, and IgA in the sera of broiler chickens, which increased by 17% ( $p<0.01$ ), 18% ( $p<0.01$ ), and 386% ( $p<0.01$ ), respectively, as compared to the control. Thus, the broiler chickens fed with Se-enriched *C. utilis* had greater immunity function than those fed diets containing other supplements or no supplements.

## DISCUSSION

Se-enriched yeast, a well-known feed additive, has already been approved by the US Food and Drug Administration as a safe source of organic Se in animal

**Table 4** – Immune functions of broiler chickens fed with different dietary supplements.

Group Supplements	A $\text{Na}_2\text{SeO}_3$	B <i>C. utilis</i>	C Se-enriched <i>S. cerevisiae</i>	D Se-enriched <i>C. utilis</i>	E Control
Immune organ index					
Thymus (%)	0.29±0.07a	0.31±0.07a	0.25±0.09b	0.34±0.10a	0.32±0.08a
Spleen (%)	0.22±0.07a	0.21±0.04a	0.29±0.08B	0.26±0.08a	0.22±0.07aA
Fabricius (%)	1.10±0.14a	1.01±0.11a	0.92±0.17B	1.01±0.16a	1.09±0.12aA
Immunoglobulin					
IgG (mg/mL)	5.37±0.19a	5.46±0.29a	4.99±0.30a	6.05±0.30B	5.17±0.27aA
IgM (mg/mL)	0.82±0.05a	0.87±0.04B	0.72±0.03B	0.94±0.03B	0.80±0.05aA
IgA (µg/mL)	29.75±2.09B	41.50±1.25B	24.75±2.59B	53.50±2.09B	11.00±1.90A

<sup>a,b,A,B</sup> In the same row, values with different lower-case letters indicate significant differences ( $p<0.05$ ), with different capital letters indicate extremely significant differences ( $p<0.01$ ), while the same letters indicate no significant differences ( $p>0.05$ ).



feedstock (Food and Drug Administration, 2000; Kieliszek *et al.*, 2015). GSH is widely distributed in yeast cells to maintain an intracellular redox environment, and a greater intracellular GSH content increases the antioxidant capacity of the yeast cells (Li *et al.*, 2004). Hence, high Se and GSH contents are essential to increase the performance of Se-enriched yeast, which, in turn, will expand the applicability of selenized yeast (Yang *et al.*, 2013). In the present study, Se-enriched *C. utilis* with high intracellular organic Se and GSH contents was used as a dietary supplement for the raising of broiler chickens. The growth performance, antioxidant capacity, and immune function of broiler chickens were also evaluated with or without dietary supplements.

Se can regulate the activities of insulin-like growth hormone and thyroid hormone growth factor, and enhance the dissimilation of fat, sugar, and protein in the body, which is beneficial to energy supply and animal growth (Burk, 2002; Fairweather-Tait *et al.*, 2011). Low dietary Se intake is associated with an increased risk in chronic diseases relating to oxidative stress (Rayman, 2000; Beck *et al.*, 2003). In this study, Se-enriched *C. utilis* was supplemented to the diet of broiler chickens, and simultaneously improved Se contents in whole blood, liver, and breast meat (Fig. 2), showing greater ability of Se accumulation, which is in accordance with those obtained in goats (Zhang *et al.*, 2018). However, no significant differences in average daily gain and drip-loss rate were found between groups of Se-enriched *C. utilis* and the control. As the drip-loss rate is an important index for measuring the quality of breast meat of broiler chickens (Phongpangan *et al.*, 2014), these results indicated that Se-enriched *C. utilis* had no remarkable contribution to improve the quality of breast meat.

The elimination of free radicals in animal bodies depends on an enzymatic system consisting of GSH-Px, CAT, and SOD, as well as a non-enzymatic system, including vitamins C, and E, and GSH (Galano & Alvarezidaboy 2011). The activities of these enzymes in the serum and tissues, as well as the GSH contents in tissues, are important indices of the antioxidant capacity of the body (Gan *et al.*, 2014). In this study, GSH-Px in the whole blood showed higher activities when Se-enriched *C. utilis* was supplemented to the diet, as compared to the control. Se is the vital cofactor of GSH-Px and GSH is the substrate (Arthur, 2000), therefore, the addition of Se-enriched *C. utilis* containing both Se and GSH favored to the increase in the activity of GSH-Px. CAT in the serum and breast meat, as well as SOD in the breast meat of broiler chickens fed with the Se-enriched *C. utilis* showed higher activities than those of the control.

Moreover, the contents of GSH in the whole blood, liver, and breast meat were also greater than that of the control. Besides, Se-enriched *C. utilis* resulted in the lowest MDA contents in serum, liver, and breast meat of broiler chickens. MDA, an end product of lipid peroxidation, is usually used as a cell index to indicate the severity of stress encountered (Koh *et al.*, 2000). The reduced MDA contents in serum and tissues of broiler chickens indicated that the Se-enriched *C. utilis* decreased the damage induced by oxygen stress. Thus, the Se-enriched *C. utilis* increased activities of GSH-Px, CAT and SOD, decreased MDA contents, resulted in much greater antioxidant capacity of broiler chickens than those of the control.

Thymus, spleen, and fabricius are important immune organs of broiler chickens. The development status and functions of these organs directly determine the chicken's immune level (Yang *et al.*, 2016). The greater the immune organ index, the greater the chicken immunity level can be expected. In this study, no significant differences in the immune organ indices were found between the dietary supplement of Se-enriched *C. utilis* and the control. However, this dietary supplement improved the levels of immunoglobulins in the serum of broiler chickens. The Se-enriched *C. utilis* resulted in the highest titers of IgG, IgM, and IgA, showing much greater immunity function than those of the control. Organic Se and GSH can increase the numbers of phagocytic cells, promote the proliferation of lymphocytes, and improve the biosynthesis of immunoglobulin, which ultimately improve the immune function of animals (Spallholz *et al.*, 1990).

Besides, other dietary supplements containing Se and/or GSH were also tested in this study. Both organic Se of Se-enriched *S. cerevisiae* and inorganic Se of Na<sub>2</sub>SeO<sub>3</sub> supplemented in the diet increased Se contents in the whole blood and liver. The supplementation of *C. utilis* cells increased GSH contents in blood, liver, and breast meat of broiler chickens, which in turn decreased MDA contents in serum and tissues. As compared to the above supplements and the control, Se-enriched *C. utilis* is the most potential feed additive for Se supplementation to improve the antioxidant capacity and immune function of broiler chickens.

In summary, the effect of Se-enriched *C. utilis* and other Se and/or GSH dietary supplements in the diet of broiler chickens was investigated in this study. Based on Se distribution, activities of GSH-Px, CAT, and SOD, GSH and MDA contents, immune organ indices, and IgG, IgM, and IgA titers in the serum, Se-enriched *C. utilis* was shown to be a good dietary supplement candidate to improve antioxidant capacity,



and immune functions of broiler chickens. This study not only indicated that the Se-enriched *C. utilis* was superior to other additives in the raising of broiler chickens but also expanded the potential applications of the selenized yeast in feed additives.

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