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Supplementing Broiler Quail Diets with Dried Egg Yolk under Heat Stress Conditions

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Abstract
This study was conducted to investigate the impact of incorporating dried egg yolk into the diet on the growth performance, carcass quality characteristics, and thigh-breast malondialdehyde value of quails (Coturnix coturnix japonica) subjected to heat stress. A total of 160 quails, 10 days old, were divided into four groups, each comprising 10 replicates with four chicks in each group. Throughout the experiment, the quails were provided with isonitrogenic and isocaloric feeds. The experimental groups were as follows: 1) Basic ration without temperature application (Control); 2) Basic ration with 1% added egg yolk, without temperature application; 3) Basic ration with temperature treatment (30 °C for 8 hours per day); and 4) Basic ration with 1% added egg yolk, along with temperature treatment (30 °C for 8 hours per day). At the end of the experiment, the highest body weight gain occurred in male and female quails which fed the diets incorporated with dried egg yolk and not exposed to heat (P<0.05). The males and females that were exposed to heat and fed with the basic ration showed the lowest feed intake, and this difference was statistically significant (P<0.05). There was no significant difference observed in the malondialdehyde (MDA) values of thigh meat on days 3 and 9, as well as in breast meat on days 3, 6, and 9 (P>0.05). However, a significant change in thigh malondialdehyde value was detected on day 6 (P<0.05). In conclusion, the addition of dried egg yolk to the diets of quails exposed to heat stress diminished the negative effects of heat stress on growth performance.

Research Article

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1. Introduction

Stress can be defined as the living organism's response to situations that disrupt normal physiological balance and functioning (Selye, 1976; Gonzalez-Rivas et al., 2020; Rostagno, 2020; Goel, 2021). The negative energy between the heat an organism releases into the surrounding environment and the heat energy it produces within is what causes heat stress (Lara and Rostagno, 2013; Rostagno, 2020). Poultry must balance the amount of heat entering their bodies with the amount of heat leaving them in order to maintain a steady body temperature (Vandana et al., 2021). It is generally agreed that the appropriate temperature range for poultry is 18–22 °C for broilers and 19–22 °C for laying hens (Vandana et al., 2021). Heat stress in poultry is classified according to the temperature and duration of exposure (Zmrhal et al., 2018). When poultry experience temperatures ranging from 27 to 38°C for a duration of 1 to 24 hours, it's termed as acute heat stress. If the exposure extends up to 7 days within the same temperature range, it's termed as sub-acute heat stress. Chronic heat stress, on the other hand, occurs when temperatures range from 38 to 50°C and persist for more than 7 days (Vandana et al., 2021). Poultry farming has many stress factors due to its nature. The most important among these stress factors is heat stress (Sahin et al., 2017; Sahin et al., 2018; Zmrhal et al., 2018; Goel, 2021; Vandana et al., 2021). The egg production, feed intake, and growth performance of poultry subjected to heat stress experience negative impacts, alongside an elevated mortality rate in the animals (Goel, 2021; Oluwagbenga and Fraley, 2023). Numerous methodologies and tactics have been devised to mitigate the adverse consequences of heat stress. Management practices such as sprinkling, ventilation, suitable housing environments, and shading are used to combat heat stress in poultry farming (Vandana et al., 2021). In addition, in order to eliminate the negative effects of heat stress, interventions are also made with various additives to poultry rations. These substances include vitamins, minerals, probiotics, and herbal products (Sahin et al., 2006). Many researchers have studied to eliminate the negative effects of heat stress by adding substances such as electrolyte balance application (Ahmad and Sarwar, 2006), sodium and chloride (Mushtaq et al., 2007), vitamin E (Sahin et al., 2001; Karami et al., 2018), A, E, zinc (Sahin et al., 2001) ascorbic acid (Ferreira et al., 2015; Karami et al., 2018) and vitamin D (Sahin et al., 2001) to the diet. Egg yolk includes various nutrients such as essential fatty acids, vitamins, minerals (Abeyrathne et al., 2022), phospholipids and antibodies (Pereira et al., 2019) as well as proteins with high biological value (Ahn et al., 1997; Anton, 2013). Chickens are animals with a significant capacity to develop antibodies (Gadde et al., 2015). These antibodies are transferred to the egg (Gadde et al., 2015; Nath and Aravindkumar, 2021). Thanks to these antibodies, animals can protect themselves from various diseases and stressful situations (Gadde et al., 2015; Pereira et al., 2019). When the structure of egg yolk is evaluated, it is seen that half of it consists of water and the remaining part consists of lipids, proteins, vitamins and minerals. It is observed that most of the lipids in the egg are in the yolk. It is seen that 62% of these lipids consist of triglycerides, 33% of phospholipids, and 5% of cholesterol (Anton, 2013; Abeyrathne et al., 2022). It is reported that the proteins in the structure of eggs have anticancer, immunomodulatory, antihypertensive, antioxidant and antibacterial properties These proteins are found in both egg whites and egg yolk (Lee et al., 2022). Additionally, egg yolk contains various color pigments known as xanthophyll carotenoids (Papadopoulos et al., 2019). Due to these properties, it is believed that the inclusion of egg yolk in the diets of poultry exposed to heat stress may reduce the harmful effects of heat stress. The present experiment was designed to assess the effects of 1% dried egg yolk supplementation to the diet on growth performance, organ weights, intestinal pH, carcass colour parameters, carcass parameters and thigh-chest malondialdehyde (MDA) levels in broiler quails raised under heat stress and thermo-neutral conditions.

2. Materiel and Methods

After Ethics Committee approval (Date: 31/05/2022 No: 2022/35), this study was conducted in accordance with the Animal Research Centre's Animal Experimentation guidelines. In the study, 160 one-day-old broiler quail (Coturnix coturnix japonica) chicks were used. The study comprised 4 groups, each consisting of 10 replicates, with 4 quail chicks in each replicate. All quails were fed a basal diet consisting of corn and soybean meal within the appropriate temperature range for rearing until day 10. The control group of the present study was fed a basal diet during the study period and housed in an environment maintained at 23 ± 2 °C after the 10th day. The egg yolk (1%) group was also fed with the basal diet supplemented with 1% dried egg yolk after day 10 while being housed in an environment with a temperature of 23 ± 2°C. The temperature (30°C) experimental group was given a basal experiment diet prepared for the study after day 10 and exposed to 30 ± 1°C for 8 h per day. Egg yolk (1%) + Temperature (30°C) group was kept at 30 ± 1°C for 8 h per day after day 10 and fed a diet supplemented with 1% egg yolk.

Table 1. Nutrient contents (%) obtained through analysis and calculation methods in the ingredients of the mixed feed formulation

Ingredients	Control	Heat (30°)	Eggyolk (1%)	Heat+Eggyolk (1%)
Sunflower oil	1.50	1.50	1.50	1.50
Corn	56.00	56.00	56.00	56.00
Soya Bean Meal (%48 CP)	33.00	33.00	33.00	33.00
Full-fat soybean	5.50	5.50	4.50	4.50
Eggyolk	0.00	0,00	1.00	1.00
DCP	1.00	1.00	1.00	1.00
Bone Flour (%30 Ca. %13 CP)	2.00	2.00	2.00	2.00
DL-Metiyonin	0.15	0.15	0.13	0.13
L-Lizin	0.20	0.20	0.22	0.22
NaCl	0.40	0.40	0.40	0.40
Vitamin and Mineral Premix	0.25	0.25	0.25	0.25
Parameters	**Values Obtained from Analysis**			
Dry Matter	89.9	89.9	89.9	89.9
Crude Protein	23.0	23.0	23.0	23.0
Crude Oil	4.40	4.40	4.69	4.69
Crude Ash	6.02	6.02	6.2	6.2
Crude Cellulose	2.82	2.82	2.80	2.80
	Calculated Chemical Composition			
ME (kcal/kg)	3016	3016	3034	3034
Ca	0.90	0.90	0.90	0.90
Available P	0.60	0.60	0.60	0.60
Na	0.19	0.19	0.19	0.19
Cl	0.28	0.28	0.28	0.28
Metiyonin+Sistin	0.90	0.90	0.90	0.90
Lizin	1.41	1.41	1.41	1.41
Treonin	0.86	0.86	0.82	0.82
Triptofan	0.30	0.30	0.30	0.30
Linoleik asit	2.38	2.38	2.33	2.33
Electrolyte Balance (mEq/kg DM)	258	258	258	258

The MDA level, a gauge of lipid oxidation, was ascertained using the thiobarbituric acid (TBA) method described by Witte et al. (1970) in quail breast and thigh flesh collected from the study. To determine milligrams per kilogram of meat, the MDA values from the spectrophotometer were corrected using a correction factor of 7.8 (Tarladgis et al., 1960).

2.1. Statistics

The data of the present study were statistically analysed using SPSS version 18.0 (SPSS, 2013). The differences between group means were evaluated using one-way analysis of variance (ANOVA). The significance of differences between groups was confirmed using the Tukey test, and significance levels were determined as P < 0.05.

3. Results

The effects of heat stress and the addition of dried egg yolk (1%) to the diet on the performance, visceral organ weights, intestinal pH, carcass color parameters, carcass parameters, and MDA values of thigh and breast muscles in quails are presented in tables (Tables 2,3,4,5,6).

Table 2. Effects of dried egg yolk supplementation on growth performance of male and female quails under heat stress

Groups	Male			Female		
	Body weight gain	Feed intake	Feed Conversion Ratio	Body weight gain	Feed intake	Feed Conversion Ratio
Control	153.7[ab]	459.6[b]	2.99[b]	197.7[ab]	633.5[ab]	3.20[ab]
Egg yolk (%1)	167.7[a]	504.2[a]	3.00[b]	216.7[a]	663.9[a]	3.06[c]
Heat (30 ºC)	147.0[b]	468.8[ab]	3.18[a]	174.4[b]	610.4[b]	3.26[a]
Egg yolk (%1) + Heat (30ºC)	160.8[ab]	489.0[ab]	3.04[b]	196.1[ab]	489.0[ab]	3.12[bc]
P	0.003	0.026	0.001	0.002	0.044	0.0001
Std. Error	2.171	5.868	0.1798	4.159	12.43	0.018

Body weight gain, feed consumption, and feed conversion ratio data of animals were determined for the methods applied to the experimental groups (Table 2). While the applied heat significantly decreased the feed intake, feed conversion ratio and body weight gain of male quails (P<0.05), the supplementation of dried egg yolk 1% to the diet significantly increased the feed intake, feed conversion ratio and body weight gain (P<0.05; Table 2). The groups that received dried egg yolk and the control group had the highest feed conversion ratio, while the groups that were subjected to heat and did not get any supplements had the lowest feed conversion ratio (Table 2). End of experiment data in female quails are as shown in Table 2. While the applied heat decreased feed intake, feed conversion ratio and body weight gain in female quails (P<0.05), dried egg yolk supplemented to the diet at 1% level caused an increase in feed intake, feed conversion ratio and body weight gain (P<0.05). In females, the best feed conversion ratio was obtained in the dried egg yolk 1% group, while the worst feed conversion ratio was obtained in the heat (30°C) group (Table 2).

Table 3. Effects of dietary dried egg yolk supplementation on internal organ weights and intestinal pH of male and female quails under heat stress

Gruplar	Male						Female					
	Gizzard	Proventriculus	Heart	Liver	Total Intestine	Intestinal pH	Gizzard	Proventriculus	Heart	Liver	Total Intestine	Intestinal pH
Control	2.44[ab]	0.33	0.80	1.72	3.50	7.16	2.59[a]	0.33[a]	0.80[a]	2.27	4.02	7.14
Egg yolk (%1)	2.40[ab]	0.29	0.88	1.51	3.15	7.17	2.20[ab]	0.31[ab]	0.81[a]	2.25	3.75	7.08
Heat(30 ºC)	2.10[b]	0.28	0.79	1.70	2.96	7.42	1.87[b]	0.27[b]	0.68[b]	2.13	3.52	7.13
Egg yolk (%1) + Heat (30 ºC)	2.61[a]	0.31	0.78	1.82	3.41	7.29	2.43[a]	0.32[ab]	0.73[ab]	2.09	3.41	7.03
P	0.022	0.083	0.127	0.217	0.579	0.221	0.007	0.049	0.032	0.941	0.494	0.805
Std. Error	0.062	0.008	0.017	0.053	0.148	0.050	0.083	0.009	0.019	0.113	0.187	0.042

The effects of dried egg yolk supplementation to male quail diets on internal organ (gizzard, proventriculus, heart, liver, total intestinal weight) weights and intestinal pH values are presented in Table 3. No differences was observed in proventriculus, heart, liver, total intestinal weights, and intestinal pH in male quails (P>0.05). A significant difference among the study groups was observed in terms of gizzard weight (P<0.05), with the dried egg yolk 1% + heat (30°C) group displaying the highest gizzard weight. Significant differences were observed in gizzard, proventriculus, and heart weights among female quails (P<0.05). On the other hand, no significant difference was detected in liver weight, total intestinal weight and intestinal pH values (P>0.05).

Table 4. Effect of dietary dried egg yolk supplementation on carcass color parameters of male quails under heat stress

Groups	Male						Female					
	Thigh L*	Thigh a*	Thigh b*	Breast L*	Breast a*	Breast b*	Thigh L*	Thigh a*	Thigh b*	Breast L*	Breast a*	Breast b*
Control	34.35	4.30	6.50	31.91	5.53[b]	7.55	36.35	4.51	7.55	31.07	6.73	8.04
Egg yolk (%1)	33.97	5.35	7.48	30.57	6.67[ab]	8.07	33.99	4.14	6.84	31.85	5.56	7.92
Heat (30 °C)	32.25	5.45	7.01	29.26	6.55[ab]	7.39	34.52	3.54	6.73	30.19	5.99	7.36
Egg yolk (%1) + Heat (30 °C)	32.94	5.03	7.25	31.18	7.25[a]	7,62	35.74	4.05	7.15	31.48	5.29	7.90
P	0.152	0.395	0.577	0.141	0.042	0.529	0.397	0.106	0.635	0.270	0.064	0.671
Std. Error	0.363	0.256	0.257	0.419	0.225	0.166	0.536	0.143	0.236	0.309	0.215	0.204

The carcasses obtained from the study groups were assessed for color parameters using the colorimetric method (Table 4). No significant difference was observed between the groups in thigh color assessment (L*, a*, b*) in male quails (P>0.05). Similarly, no significant difference was determined in breast L* and breast b* (P>0.05), while a statistically significant change was detected in breast a* (P<0.05). The color parameters of thigh and breast in female quails were assessed using the calorimetric method. No significant differences were observed in thigh and breast color parameters (L*, a*, b*) in female quails (P>0.05).

Table 5. Effects of dietary dried egg yolk supplementation on carcass parameters of male quails under heat stress

Groups	Male			Female		
	Carcass Yield	Hot Carcass	Cold Carcass	Carcass Yield	Hot Carcass	Cold Carcass
Control	62.62	130.23	122.13[ab]	60.45	145.15	134.09
Egg yolk (%1)	60.97	128.25	124.56[a]	58.63	136.86	129.94
Heat(30 °C)	61.25	123.60	109.04[b]	58.17	137.06	122.09
Egg yolk (%1) + Heat(30 °C)	61.33	124.53	114.39[ab]	56.98	138.29	128.76
P	0.390	0.589	0.018	0.470	0.650	0.402
Std. Error	0.360	1.900	2.033	0.770	2.579	2.473

The effects of dietary dried egg yolk supplementation on carcass yield, hot carcass weight and cold carcass weight are shown in Table 5. There was no significant difference among the groups in terms of carcass yield and hot carcass weight (P>0.05). However, the group receiving dried egg yolk showed the highest cold carcass weight (P<0.05). Similarly, no difference was detected between the female groups in terms of cold carcass, hot carcass and carcass yield weights (P>0.05).

Table 6. Effect of dirtay dried egg yolk supplementation on the diets of quails under heat stress on the MDA value of thigh and breast meat on the 3rd, 6th, and 9th day

	MDA					
Gruplar	Thigh 3	Thigh 6	Thigh 9	Breast 3	Breast 6	Breast 9
Control	0.1253	0.0807[a]	0.1308	0.1250	0.0715	0.1198
Egg yolk (%1)	0.1168	0.0485[ab]	0.1358	0.1290	0.0678	0.1313
Heat (30 ºC)	0.1392	0.0608[ab]	0.1427	0.1380	0.0603	0.1628
Egg yolk (%1) + Heat (30ºC)	0.1243	0.0678[b]	0.1310	0.1325	0.0753	0.1333
P	0.775	0.010	0.812	0.936	0.731	0.559
Std. Error	0.00719	0.00389	0.00462	0.00679	0.00457	0.01049

MDA levels in thigh and breast meat were analyzed on the 3rd, 6th, and 9th days after slaughter as a result of the addition of egg yolk to quail diets. The effects of heat stress with the addition of egg yolk to quail diets were not detected on MDA values on thigh and breast meat on thigh 3, thigh 9, breast 3, breast 6, and breast 9 between the study groups (P>0.05). There was a significant difference between the groups in terms of thigh MDA values on day 6 (P<0.05). The lowest MDA value was found in the egg yolk (1%) group.

4. Discussion

In recent years, many natural and synthetic supplements (Sahin et al., 2001; Ahmad and Sarwar, 2006; Mushtaq et al., 2007; Karami et al., 2018; Mehaisen et al., 2019; Pereira et al., 2019) have been used to alleviate the negative effects of heat stress on poultry performance and maintain maximum production at high environmental temperatures. In the present study, feed conversion ratio, feed intake and body weight gain were negatively affected in both male and female animals exposed to heat stress (Table 2). The best results were obtained in animals not exposed to heat stress and received only dried egg yolk (1%) supplementation, while the data of animals exposed to heat stress and receiving dried egg yolk (1%) supplementation were improved (Table 2). Egg yolk is nutrient-rich in proteins, vitamins, minerals, and immunoglobulins (IgY) as well as essential fatty acids (Anton, 2013; Pereira et al., 2019; Xiao et al., 2020; Abeyrathne et al., 2022). Heat stress negatively affects egg production, feed consumption, and growth performance in poultry, and this stress condition not only causes low productivity in animals but also negatively affects health (Anton, 2013; Pereira et al., 2019; Xiao et al., 2020; Abeyrathne et al., 2022). The improvement obtained with the addition of 1% dried egg yolk to the diet in groups exposed to heat stress may be attributed to the capacity of essential fatty acids in the yolk structure to reduce heat stress. Because 65% of the dry matter of egg yolk consists of lipids (Xiao et al., 2020). As fats are metabolized, they raise body temperature less than proteins and carbohydrates (Attia and Hassan, 2017; Wasti et al., 2020). Attia and Hassan (2017) and Ghazalah et al. (2008) previously showed that the effects of heat stress were reduced by adding different amounts of fat to the diet. A similar effect may have been obtained due to the fatty acids in the structure of egg yolk. In this sense, similar results were obtained with our present study. Because the animal exposed to the hot environment spends most of its time performing the actions required to cool the body (Mack et al., 2013) rather than eating.

Table 3 shows the internal organ weights and intestinal pH values of male and female quail of the experimental groups. According to the experiment results, gizzard weight of male quails showed a difference depending on the treatments (P<0.05), while proventriculus, heart, liver, total intestinal weight, and intestinal pH did not change (P>0.05). In males, the highest gizzard was obtained in the group that was exposed to heat stress and received dried egg yolk supplementation at the same time (P<0.05). The lowest gizzard weight was obtained in the group exposed to temperature stress without any supplementation (P<0.05). There was no significant difference among the female quail groups in terms of liver, total intestinal, heart weights and intestinal pH values (P>0.05). There were differences in gizzard, proventriculus and heart weights (P<0.05). The highest gizzard, stomach with gland, and heart weights were obtained in the control groups (P<0.05). The lowest gizzard, proventriculus, and heart weights were found in groups exposed to heat stress. In many previous studies, negative side effects of heat stress on internal organ weights have been mentioned (Onderci et al., 2005; Sahin et al., 2009; Lara and Rostagno, 2013; Sahin et al., 2013; He et al., 2019; Chang et al., 2020). The increase in organ weights may be an indication of the development of the organism. As the organism grows, the internal organs grow within their physiological limits. Adverse conditions during the growth period can affect the development of internal organs. In the present study, the supplementation of dried egg yolk to the diet of quails exposed to heat stress may have reduced the side effects of heat stress due to the properties of egg yolk (Ahn et al., 1997; Anton, 2013; Gadde et al., 2015; Pereira et al., 2019; Abeyrathne et al., 2022).

Meat color in poultry and other animals may vary depending on age at slaughter, stress at the time of slaughter, genetic factors, gender, and especially the effect of nutrition (Warner, 2014). The color of meat is determined by factors such as the concentration of myoglobin on the surface of the meat, its chemical structure, texture, and light scattering (Warner, 2014; Kong et al., 2023). Meat color is an important criterion for consumer preferences (Warner, 2014; Kong et al., 2023). In the L* a* b* evaluation system, L* is used to denote the brightness, a* the redness coordinate, and b* the yellowness coordinate (Le Bihan-Duval et al., 2008; Warner, 2014; Salueña et al., 2019). The quality of thigh and breast meat was measured and evaluated using the L*a*b* colour correction system (Table 4). In the color criteria, only in the male quail group, breast a* was higher than the others (P<0.05). No difference was detected in the other groups (Table 4). The high a* value indicates that the redness of the meat increases (Warner, 2014; Kong et al., 2023). In that study, a difference was found in terms of breast a* in male quails (P<0.05). There was no significant difference in other data (P<0.05). Heat stress causes glycogen to be converted into lactic acid in muscles faster than normal, causing a* value to decrease, Hao and Gu (2014) and Zaboli et al. (2019) studies are similar to our current study.

Carcass yield, hot carcass, and cold carcass data were measured in male and female quails (Table 5). Experimental results showed that the addition of dried egg yolk to the diet of quails under heat stress increased cold carcass yield in male quails (P<0.05). Considering the cold carcass data, both male and female quails were found to be low in the group exposed to temperature stress. This can be linked to the rapid anaerobic glycolysis effect of heat stress on muscles (Zaboli et al., 2019; Vandana et al., 2021). Because in muscles exposed to heat stress, it causes glycogen to convert to lactic acid faster than normal (Zaboli et al., 2019). This rapid transformation can cause more water loss in the carcass in the time after slaughter (Hao and Gu, 2014; Zaboli et al., 2019). These studies (Hao and Gu, 2014; Zaboli et al., 2019) are similar to our current study.

One byproduct of polyunsaturated fatty acid peroxidation in cells is MDA. MDA increases as a result of the organism's increased production of free radicals. It is widely acknowledged that elevated MDA levels indicate oxidative stress. In the current study, a decrease was detected in MDA values on the 6th day of thigh when compared to the remaining groups. On the other days, although there was no statistical difference in MDA levels, the highest MDA values were found in the group exposed to heat stress and not receiving any supplementation. Sahin et al. (2006) reported a decrease in liver MDA levels in their study in which vitamin E and zinc picolinate were added to the diet. Sahin et al. (2002) found that Vitamin E and C decreased MDA levels in quails exposed to heat stress. Onderci et al. (2003) reported that zinc supplementation decreased serum MDA levels in heat stress. Sahin et al. (2002) egg is a rich food containing various proteins such as ovalbumin, ovotransferrin, and fosvitin as well as lipids such as phospholipids. It also contains many micronutrients such as vitamin E, vitamin A, selenium, and carotenoids, which makes the egg a food with antioxidant properties. These antioxidants can reduce cellular damage by fighting free radicals (Nimalaratne and Wu, 2015). In the present study, dried egg yolk added to the diet may have prevented the increase in MDA due to these effects.

5. Conclusions

The problem of broken eggs is an important economic loss in laying hens. Broken eggs can be evaluated as feed additives to reduce the adverse impacts of heat stress on poultry by adding dried eggs to poultry diets. Thermostress is one of the major environmental stress factors in poultry. This situation causes various negativities in terms of both yield and meat quality in poultry. There are very few studies on the inclusion of egg yolk obtained from broken eggs in the diet. There is a need for more extensive and comprehensive studies on this subject.

Declaration of Author Contributions

The authors declare that they have contributed equally to the article. All authors declare that they have seen/read and approved the final version of the article ready for publication.

Declaration of Conflicts of Interest

All authors declare that there is no conflict of interest related to this article.

Ethical Committee Approval

After Ethics Committee approval (Date: 31/05/2022 No: 2022/35), this study was conducted in accordance with the Animal Research Centre's Animal Experimentation guidelines.

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