UNIVERSITI PUTRA MALAYSIA

EFFECTS OF DIETARY SUPPLEMENTATION OF GLUTAMINE AND GLUTAMIC ACID ON PERFORMANCE AND STRESS RESPONSE TO HIGH TEMPERATURE IN BROILER CHICKENS

OLUBODUN JOSHUA OMOLAYO

ITA 2015 9

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By

OLUBODUN JOSHUA OMOLAYO

Thesis Submitted to the School of Graduate Studies, Universiti Putra Malaysia, in Fulfillment of the Requirement for the Degree of Master of Science

July 2015

DEDICATION

This work is dedicated to the Almighty God who gave me life, and help throughout this programme may His name be praised for ever. I also dedicate this thesis to the loving memory of my late brother Timothy Oluwabusayo Olubodun, may his soul continue to rest in the bosom of Abraham till we meet to part no more. We miss you.

Abstract of thesis presented to the senate of Universiti Putra Malaysia in fulfillment of the requirement for the degree of Master of Science

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Chairman: Prof. Zulkifli Idrus, PhD

Institute: Institute of Tropical Agriculture

Glutamine (Gln), a conditionally essential amino acid has been documented to aid animals under stressful condition which include injury, weaning, infection and overcrowding. Reports on the following functions of Gln have been inconsistent hence, the need to ascertain this claims and especially under high temperature. Experiments were conducted to investigate the effects of dietary supplementation of glutamine (Gln) and glutamic acid (Glu) on performance and stress response to high temperature in broiler chicken. In experiment 1, day-old (day 1) commercial broiler chicks were fed (i) basal diet (control), (ii) basal diet + 0.5% Gln+Glu or (iii) basal diet + 1% Gln+Glu from 1 to 42 days of age under the hot and humid tropical environment. AminoGut (Gln+Glu) is a commercial dietary supplement containing a mixture of L-glutamine (Gln) and L-glutamic acid (Glu). Weight gain and FCR during the starter (day 1-21) and overall (day 1-42) periods improved linearly and quadratically with Gln+Glu supplementation when compared to control. Supplementing birds with Gln+Glu significantly reduced overall mortality rate. At 21 and 42 days of age, intestinal (duodenum and ileum) villi height and crypt depth showed both linear and quadratic positive responses to Gln+Glu supplementation. Intestinal amylase activity increased linearly and quadratically on day 21, and linearly only on day 42. It was concluded that Gln+Glu supplementation was beneficial in improving the growth performance and survivability of broiler chickens under the hot and humid tropical environment. Experiment 2 was conducted to investigate the effects of Gln+Glu supplementation on growth performance and physiological stress response in broilers exposed to cyclic heat stress. Day-old commercial broiler chicks were kept in environmentally controlled rooms and fed basal diet (control) or basal diet + 0.5% Gln+Glu from 1 to 21 days of age. From day 22-42, chicks in one chamber were exposed to 34±1^{o}C for 5 hours daily while the other group of chicks remained under 24^{o}C throughout. Weight gain and FCR were significantly improved during the starter (day 1-21) and overall (day 1-42) periods with Gln+Glu supplementation when compared to control. Supplementing

birds with Gln+Glu also significantly reduced mortality rate in birds during the heat challenge period. Provision of Gln+Glu to the heat stressed birds resulted in higher heat shock protein (HSP) 70 expression and serum acute phase protein ovotransferrin concentration (OVT). In conclusion, the beneficial effect of Gln+Glu supplementation on performance and survivability of broilers under heat stress could be attributed to enhanced HSP 70 and OVT synthesis.

Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia sebagai memenuhi keperluan untuk Ijazah Master Sains

KESAN PENAMBAHAN GLUTAMINE DAN GLUTAMIC ACID TERHADAP PRESTASI AYAM PEDAGING DAN TINDAK BALASNYA KEPADA TEKANAN HABA TINGGI

Oleh

OLUBODUN JOSHUA OMOLAYO

Julai 2015

Pengerusi: Prof. Zulkifli Idrus, PhD

Fakulti: Institut Pertanian Tropika

Glutamine (Gln),iaitu asid amino perlu, telah didapati dapat membantu haiwan yang di dalam keadaan stress, samada disebabkan oleh kecederaan, dipisahkan dengan ibu, jangkitan penyakit atau ditempatkan dalam kawasan yang terlalu sempit. Walaubagaimanapun, laporan-laporan mengenai fungsi Glutamine tersebut adalah tidak konsisten. Oleh itu, adalah perlu untuk membuktikan perkara ini, terutamanya untuk keadaan apabila ternakan dipelihara dalam persekitaran yang bersuhu tinggi. Kajian-kajian telah dijalankan untuk mengkaji kesan pemberian makanan yang dicampurkan dengan glutamine (Gln) dan glutamic acid (Glu) terhadap prestasi ayam dan tindak balasnya kepada suhu yang tinggi. Dalam eksperimen 1, ayam komersial yang berusia satu hari telah diberi makan samada (i) makanan biasa (control), (ii) makanan biasa + 0.5% Gln+Glu, atau (iii) makanan biasa +1% Glu+Gln, bermula dari hari pertama hingga hari ke-42, dan dipelihara dalam persekitaran tropika yang panas dan lembap. AminoGut (AG) adalah makanan tambahan komersial yang mengandungi campuran L-glutamine dan L-glutamic acid (Gln+Glu). Kenaikan berat badan ayam dan FCR ketika fasa permulaan (hari 1-21) dan dalam waktu keseluruhan waktu kajian (hari 1-42) adalah lebih baik apabila ayam diberi makanan yang dicampurkan dengan Gln+Glu, berbanding apabila diberi makanan biasa sahaja (control). Kadar kematian ayam secara keseluruhan juga berkurang apabila ayam diberi makanan yang mengandungi campuran Gln+Glu. Aktiviti enzim amylase usus juga telah meningkat secara linear dan kuadratik pada hari ke-21, manakala ia meningkat hanya secara linear pada hari ke-42. Dapat disimpulkan bahawa penambahan Gln+Glu dalam makanan memberi manfaat dalam penambahbaikan prestasi pertumbuhan dan kemampuan ayam untuk meneruskan kelangsungan hidup, apabila dipelihara dalam persekitaran tropika yang panas dan lembap. Eksperimen 2 dijalankan untuk mengkaji kesan penambahan Glu+Gln terhadap prestasi pertumbuhan dan respon fisiologi ayam apabila didedahkan kepada tekanan haba berjadual. Ayam berusia satu hari dipelihara dalam bilik suhu terkawal dan diberi makanan biasa (control) atau makanan biasa +0.5% Gln+Glu mulai hari 1 hingga 21. Dari hari 22-42, ayam di dalam satu bilik tertutup didedahkan kepada suhu 34±1^{o}C selama 5 jam setiap hari, manakala kumpulan-kumpulan ayam lain dipelihara kekal di bawah suhu 24^{o}C, sepanjang masa kajian. Berbanding diberi

makanan biasa sahaja, penambahan Gln+Glu dalam makanan telah memberi kesan baik kepada kenaikan berat badan dan FCR, untuk kedua-dua fasa permulaan (hari 1-21) dan keseluruhan (hari 22-42). Kadar kematian ayam yang dipelihara dalam keadaan bersuhu tinggi juga telah berkurang apabila ayam diberi makanan yang dicampurkan dengan Gln+Glu. Penambahan Gln+Glu dalam makanan ayam yang didedahkan kepada tekanan haba telah menyebabkan kenaikan 'heat shock protein' (HSP) 70 dan serum 'acute phase protein ovotransferrin' (OVT). Kesimpulannya, penambahan Glu+Gln dalam makanan ayam amat bermanfaat kepada prestasi dan kemampuan ayam untuk meneruskan kelangsungan hidup apabila dipelihara dalam tekanan haba, dengan cara peningkatan pengeluaran HSP 70 dan OVT.

ACKNOWLEDGEMENTS

This piece of work wouldn't have been possible without the help of the Father, Son and the Holy Ghost. My profound gratitude goes to God Almighty, the one who was, who is and who is to come, the author and the finisher of my faith for His faithfulness and love over me, my family, my supervisor and his family and over my team members, may His name be praised for ever.

My sincere appreciation goes to my supervisor, Prof. Dr. Zulkifli Idrus for his patience, guidance, fatherly love and dedication shown to me during the course of my study, may the Lord prosper your ways for ever.

I would like to appreciate my co- supervisors Prof. Dr. Mohd Hair Bejo, Assc. Prof.Dr. Azhar Kasim for their contributions during this programme God bless you. I am indebted to Dr. Abdoreza soleimani Reza (Research Fellow), whose open hands welcome me at all times during this programme. I appreciate my group members Zulkifli, Pardis, Majid, Ahmed, Fatin, El-muataz and Nora for their support. I also appreciate every staff of Institute of Tropical Agriculture, Animal Science Laboratory; Serology Laboratory, Faculty of Veterinary Medicine; Meat Science Laboratory, Faculty of Agriculture; Ladang Two staffs for their support throughout my field and laboratory work, God bless you all.

On a final note, my thanks go to my caring and lovely parents for all their efforts to make me somebody in life and their words of encouragement which kept me going. I will like to appreciate my siblings: Sanmi, Sunday, Funbi, a special friend Mercy and my extended family members for their prayers, encouragement and support at all times, May the Lord increase you forever. To my Pastors, Dr.& Mrs U.I. Aloysius, Engr. Esan, Rowland, Michael, Festus, Paul, my friends Ola, Michael, Busola and other M.I.C.F and R.C.C. families for their prayers , financial, spiritual and moral support may your generations never lack helpers.

To God alone be adoration, glory and honour forever and ever for the completion of this programme. Amen.

This thesis was submitted to the Senate of Universiti Putra Malaysia and has been accepted as fulfillment of the requirement for the degree of Master of Science. The members of Supervisory Committee were as follows:

Zulkifli Idrus, PhD
Professor
Institite of Tropical Agriculture
Universiti Putra Malaysia
(Chairman)

Mohd. Hair Bejo, PhD
Professor
Faculty of Veterinary Medicine
Universiti Putra Malaysia
(Member)

Azhar Kasim, PhD
Associate Professor
Faculty of Agriculture
Universiti Putra Malaysia
(Member)

……..

BUJANG KIM HUAT, PhD
Professor and Dean
School of Graduate Studies
Universiti Putra Malaysia

Date:

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Name and Matric No.: Olubodun Joshua Omolayo GS29885

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Name of Member of Supervisory Committee:	_____	Name of Member of Supervisory Committee	_____

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LIST OF ABBREVIATIONS

AG	AminoGut
AGP	Alpha -1- acid glycoprotein
API	Acute phase protein index
APP	Acute phase protein
APR	Acute protein response
ATP	Adenosine triphosphate
^{o}C	Degree centigrade
CD	Crypt depth
CORT	Corticosterone
CP	Ceruloplasmin
CPS	Carbamoyl phosphate synthetase
CRH	Corticotropin-releasing hormone
CRP	C-reactive protein
D	Day
DOA	Day of age
DNS	Dinitrosalicylic acid
DPL	Dietary protein level
FCR	Feed conversion ratio
FI	Feed intake
GIT	Gastrointestinal tract
Gln	Glutamine
Glu	Glutamic acid
H	Hour
HLR	Heterophil and lymphocyte ratio
HPA	Hypothalamus pituitary axis
HS	Heat stress
HSP	Heat shock protein
kCal	Kilocalorie
kg	Kilogram
ME	Metabolizable Energy
mL	Millimetre
N	Nitrogen
Ng	nanogram
Nm	Nanometer
OVT	Ovotransferrin
SAP	Serum amyloid p-component
TCZ	Thermoneutra comfort zone.
UA	Uric acid
V	volt
VH	Villi height
WG	Weight gain
µg	microgram
µL	microlitre

CHAPTER 1

GENERAL INTRODUCTION

Feed additives are commonly used in the poultry industry to enhance growth performance, health and well-being. Supplementing poultry diets with specific functional amino acids have been widely investigated (Windisch et al., 2007). Recently, there is a lot of interest in glutamine, a non-essential amino acid, as a feed supplement for poultry.

L-glutamine (Gln), an abundant amino acid in the body, has been traditionally considered a non-essential amino acid but is now regarded as a conditionally essential nutrient in animals under stressful conditions such as infection, injury, weaning and heat stress(Dai et al., 2011; Lara and Rostagno, 2013). Gln is an important precursor for the synthesis of amino acids, nucleotides, nucleic acids, amino sugars, proteins, and many other biologically important molecules (Souba, 1990). Gln is a major energy substrate for rapidly dividing cells (including enterocytes and lymphocytes) and other cells, providing ATP for intracellular protein turnover, nutrient transport through the plasma membrane, cell growth and migration, as well as the maintenance of cell integrity (Li et al., 2007). Gln has so many functions in both animal and man, part of which include; standing against bacteria attack and mucosa structure maintenance (Khan et al., 1999). Inoue et al. (1993) reported that Gln supplementation stimulates gut mucosa proliferation in rats. It also helps in maintaining gut integrity (Menconi et al., 2013).

Dietary amino acids in excess of the amounts needed for growth and maintenance of protein turnover are preferentially degraded over carbohydrates and lipids since animals cannot store these excess amino acids (Campbell, 1991). Metabolism of carbohydrates and lipids forms essentially a single waste product, carbon dioxide, while amino acid degradation also releases nitrogen as ammonia, a relatively toxic molecule (Cooper and Plum, 1987). Whereas aquatic species can excrete ammonia directly, semi aquatic and terrestrial animals detoxify ammonia, at a considerable energetic cost, through the synthesis of urea (mammals) or uric acid (birds) or urea and uric acid (reptiles).

According to Wu et al. (2007), Gln participates in many key metabolic processes such as protein synthesis, gluconeogenesis, inter-organ nitrogen transfer, nucleic acid biosynthesis and immune response. Gln supplementation also enhances anti-oxidative function and cell proliferation in the small intestine (Sakamoto et al., 2014). Many benefits have been observed due to Gln supplementation in the diet of humans and rats. However, little research has been done in livestock and poultry (Dai et al., 2009). Dietary Gln supplementation as reported by Wu et al. (2007), prevented jejunal atrophy in early-weaned piglets during the first week post-weaning and improved growth performance. Yi et al. (2001) reported that supplementing 1% Gln improved weight gain (WG) and feed conversion ratios (FCR) of turkey poults. Additionally, providing 1% Gln to chickens increased villus height of the small intestines (Murakami et al., 2007) and improved resistance to *Eimeria maxima* challenges (Yi et al., 2005).

L-glutamic acid or glutamate is produced from Gln by glutaminase in the small intestine (Wu, 1998). Dietary Gln is a specific precursor for the intestinal synthesis of glutathione, arginine and proline (Reeds et al., 1996; Wu and Morris 1998). Thus,

supplementation of Gln in animal diet during stressful conditions is of critical importance in intestinal metabolism and physiology (Shizuka et al., 1990; Wu et al., 2004; Wang et al., 2009). Dai et al. (1999, 2011) reported that dietary Gln supplementation improved broiler performance during heat stress conditions.

Physiological stress such as high temperature could cause protein folding, protein damage, cellular injury and even death (Wischmeyer, 2002). Cells have developed some protective mechanisms to promote their survival during heat stress (HS) by synthesis of a group of polypeptide family of proteins called heat shock proteins (HSP). Gln has been documented to enhance heat tolerance (Nissim et al., 1993) and feeding of Gln was reported to increase HSP 70 expression. HS results in activation of the hypothalamic-pituitary-adrenal (HPA) axis activity and increases circulating corticosterone and heterophil and lymphocytes ratio (HLR), the common indices of physiological stress in avian species (Altan et al., 2003; Costa-Pinto and Palermo-Neto, 2010). It is well established that heat stress can reduce performance parameters such as feed intake, weight gain and feed efficiency in chickens (Donkoh, 1989; Siegel, 1995; Dai et al., 2009). According to Mitchell and Carlisle, (1992), and Quinteiro-Filho et al. (2010), HS reduced intestinal villi height and the wet and dry weights of jejunum. Teeter and Belay (1996) reported that chicken tends to manage HS by increasing the physiological processes responsible for heat dissipation including elevated body temperature, panting and respiratory alkalosis and the physiological processes directed at decreasing heat production. Another group of proteins that come to play during physiological stress is acute phase protein.

Acute phase proteins (APP) are a group of blood proteins that change in concentration when animals were subjected to external and internal challenges such as infection, tissue injury and inflammation.(Murata et al., 2004). These set of proteins' concentrations may increase (positive acute phase proteins) up to 25% and may decrease (negative acute phase proteins) during inflammatory conditions. There is now evidence that stress may also induce APP reaction in poultry. Recent work by Zulkifli et al. (2014) showed that administration with exogenous corticosterone elevated serum levels of acute phase proteins (APP) in broiler chickens. Overcrowding stress has been shown to increase serum levels of APP alpha-1 acid glycoprotein, ceruloplasmin and ovotransferrin in broilers (Shakeri et al., 2014). According to Tirziu (2009), APP works by natural defense mechanism networks through rehabilitating homeostasis, decreasing damages intensity, amplifying resistance, and opsonization.

Many benefits of Gln supplementation in the diets of humans (Medina, 2001; Newsholme, 2001; Andrew and Griffiths, 2002), pigs (Kitt et al., 2002), rats (Inoue et al., 1993), and mice (Adjei et al., 1994) had been reported. There is, however, a dearth of documented work on L-glutamine and L-glutamic acid in poultry. The present work focused on evaluating the effect of Gln+Glu supplementation on performance, intestinal morphology, and stress response of broiler chickens under heat stress condition.

1.1 Justification

Recent studies in poultry investigated the effect of supplementing Gln alone and the recommended duration of supplementation is not clear. Bartell and Batal (2001) reported that feeding 1% Gln for 21 days significantly improved the body weight and intestinal villi length in chickens. On the contrary, Ebadiasl (2011) reported that

feeding 1% Gln for 28 days has no significant effect on the performance of broiler chickens. Gln+Glu supplementation works in the intestine and had direct effects on enterocytes (Wu, et al., 2015). However, recent work by Shakeri et al. (2014) showed that supplementing broilers with Gln+Glu for 1-21 and 1-42 days had negligible effect on growth performance but improved the survivability rates of heat-stressed broiler chickens raised at different stocking densities. Hence, it is necessary to ascertain the effect of Gln+Glu supplementation on broiler chickens under heat stress condition. This work will ascertain appropriate dosage of Gln+Glu for optimal performance in broiler chicken and also study the works of Gln+Glu in the system of broiler as it concern heat stress condition.

1.2 General objective

To investigate the effect of dietary supplementation of Gln+Glu on performance and physiology of broiler chickens raised under hot environment (temperature higher than thermoneutral zone 24^{o}C and relative humidity 60-80%).

i. To determine the effect of dietary supplementation of Gln+Glu on the performance, intestinal morphology and amylase activity in broiler chickens under the hot humid tropical condition

ii. To investigate the effect of Gln+Glu supplementation during the starter period on heat shock protein (HSP) and acute phase protein (APP) reaction to heat stress in broiler chickens.

REFERENCES

Abraham, P., Leftheriotis, G., Desvaux, B., Saumet, M., Saumet, J. L., (1994). Diameter and blood velocity changes in the saphenous vein during thermal stress. Eur. J. Appl. Physiol., 69:305–308.

Adjei, A.A., Y. Matsumoto, T., Oku, Y.H. and Yamamoto., S., (1994). Dietary arginine and glutamine combination improves survival in septic mice. Nutr. Res., 14: 1591-1599.

Al-Aqil A., and Zulkifli, I., (2009). Changes in heat shock protein 70 expression and blood characteristics in transported broiler chickens as affected by housing and early age feed restriction. Poult. Sci., 88:1358–1364

Altan, A., Pabuascuoaylu, A., Altan, A., Konyalioaylu, S., and Bayraktar, H., (2003). Effect of heat stress on oxidative stress, lipid peroxidation and some stress parameters in broilers. Br. Poult. sci., 44: 545-550.

Alverdy, J. C., (1990). Effects of glutamine-supplemented diets on immunology of the gut. J. Pen.- Parenter. Enter., 14:109S-113S.

Alverdy, J., Aoys, E., Weiss-Carrington, P., Burke, D., (1992).The effect of glutamine-enriched TPN on gut immune cellularity. J. Surg. Res., 52:34-38.

Ambrogelly, A., Palioura, S., and Soll, D., (2007).Natural expansion of the genetic code. Nat. Chem. Biol., 3: 29–35.

Andrews, F.J and Griffiths R.D., (2002). Glutamine: essential for immune nutrition in the critically ill. Br. J. Nutr., 51:3-8.

Arjona, A. A., D. M. Denbow, and W. D. Weaver Jr., (1988). Effect of heat stress early in life on mortality of broiler exposed to high environmental temperatures just prior to marketing. Poultry Sci. 67: 226-231.

Arthington, J., Eicher, S., Kunkle, W.E. and Martin, F.G., (2003). Effect of transportation and commingling on the acute-phase protein response, growth, and feed intake of newly weaned beef calves. J. Anim. Sci., 81, 1120-1125.

Ashkenazi, J., Carpentier, Y.A., Michelsen, C.B., Elwyn, D.H., Furst, P., Kantrowitz, L.R., Gump, F.E. and Kinney, J.M., (1980). Muscle and plasma amino acids following injury: influence of intercurrent infection. Annal. Surg., 192: 78–85.

Avellaneda, Y., Hernandez, J., Ariza, C. and Afanador, T., (2008). Effect of L-Glutamine and L-Glutamate (Aminogut) supplementation on the early growth of broilers. Rev. Med. Vet.Zoot. 55: 77-90.

Awad, E. A., Fadlullah, M., Zulkifli, I., Soleimani, A. F., Loh, T. C., (2014). Amino acids fortification of low-protein diet for broilers under tropical climate: ideal essential amino acids profile. Ital J Anim Sci., 13:270-274

Azad, M. A. K., Kikusato, M., Maekawa, T., Shirakawa, H., Toyomizu, M., (2010).Metabolic characteristics and oxidative damage to skeletal muscle in broiler chickens exposed to chronic heat stress.Comparative Biochem and Physiol. 155: 401-406.

Baghshani, H., Nazifi, S., Saeb, M., and Saeb, S., (2010).Influence of road transportation on plasma concentrations of acute phase proteins, including fibrinogen, haptoglobin, serum amyloid A, and ceruloplasmin, in dromedary camels (Camelus dromedaries). Com. Clin. Pathol., 19:193-198.

Barbe, M.F., Tytell, M.D., Grower, D.J., and Welch, W.J., (1988). Hyperthermia protects against light damage in rat retina. Sci., 241: 1817–1820.

Bartell, S., and Batal, A., (2007).The effect of supplemental glutamine on growth performance, development of the gastrointestinal tract, and humoral immune response of broilers.Poult. Sci., 86:1940-1947.

Becker, B. F., (1993). Towards the physiological function of uric acid. Free Radical Biol. Med., 14: 615–631.

Bayne, C.J. & Gerwick, L. (2001).The acute phase response and innate immunity of fish. Dev. Comp. Immuno., 25:725-743.

Beloor, J., H. K. Kang, Y. J. Kim, V. K. Subramani, I. S. Jang, S. H. Sohn and Y. S. Moon, (2010). The effect of stocking density on stress related genes and telomeric length in broiler chickens. Asian-Aust. J. Anim. Sci.,4 : 437 – 443.

Bernfeld, P., (1955). Amylases, alpha and beta Method. Enzymol., 1: 149-158.

Berres, J., Vieira, S. L., Favero, A., Freitas, D. M., Pena, J. E .M., Nogueira, E. T., (2011). "Digestible valine requirements in high protein diets for broilers from twenty-one to forty-two days of age."Ani. Feed Sci. Tech., 165: 120-124.

Bezerra RM, Perazzo FG, Santos RA (2013).Glutamic acid and protein reduction for broilers and commercial laying hens. Braz. J. Appl. Tech. Agric. Sci., 6:101-109.

Blikslager, A. T., M. C. Roberts, J. M. Rhoads, R. A. Argenzio. (1997). Is reperfusion injury an important cause of mucosal damage following porcine intestinal ischemia? Surg., 121: 526–534.

Bode B. P., (2001). Recent molecular advances in mammalian glutamine transport. J Nutr., 131: 2475S-85S.

Brian, I. L., andSouba, W. W., (2000).Glutamine. World J. Surg., 24: 1503–1513.

Burkholder, K. M., Thompson, K. L., Einstein, M. E., Applegate, T. J., and Patterson, J. A., (2008). Influence of stressors on normal intestinal microbiota, intestinal morphology, and susceptibility to Salmonella enteritidis colonization in broilers. Poult. Sci., 87:1734–1741.

Burch, H. B., Narins, C., Chu, C., Fagiolo, S., Choi, S., McCarthy, W., and Lowry, O. H., (1978).Distribution along the rat nephron of three enzymes of gluconeogenesis in acidosis and starvation. Am. J. Physiol., 235: 246–253.

Burrin D. G., and Stoll B., (2009).Metabolic fate and function of dietary glutamate in the gut. Am. J. Clin. Nutr ., 90:850–856.

Campbell J. W. (1991).“Excretory nitrogen metabolism” in Environmental and Metabolic Animal Physiology. Comparative Animal Physiology, 4th Edn., ed. Prosser C. L., editor. (New York: Wiley-Interscience;), 277–324.

Cawthon, D., Mcnew, R., Beers, K. W., Bottje, W. G., (1999). Evidence of mitochondrial dysfunction in broilers with pulmonary hypertension syndrome (Ascites): effect of t-butyl hydroperoxide on hepatic mitochondrial function, glutathione, and related thiols.Poult. Sci., 78:114-124.

Chaudhry, F. A., Reimer, R. J., and Rober, H. E., (2002). The glutamine commute take the N line and transfer to the A. J. Cell Biol., 157: 349–355.

Chokshi, N.K., Guner, Y.S., Hunter, C.J., Upperman, J.S., Grishin, A., Ford, H.R., (2008).The role of nitric oxide in intestinal epithelial injury and restitution in neonatal necrotizing enterocolitis. Semin.Perinatol. 32: 92-99.

Chow, A and Zhang, R., (1998).Glutamine Reduces Heat Shock–Induced Cell Reath in rat intestinal epithelial cells. J. Nutr., 128: 1296-1301

Cooper J. L., Plum F. (1987).Biochemistry and physiology of brain ammonia. Physiol. Rev. 67, 440–519.

Costa-Pinto, F. A. D. and Palermo-Neto J., (2010).Neuroimmune interactions in stress. Neuro, 17: 196-199.

Cravener, T. L., Roush W. B. and. Mashaly M. M., (1992).Broiler production under varying population-densities.Poult. Sci. 71:427-433.

Cray, C., Zaias, J. and Altman, N.H., (2009). Acute phase response in animals: A review. Comp. Med., 59: 517–526.

Curi, R., Newsholm, P., Procopio, J., Lagranha, C., Gorjão, R., and Pithon-Curi, T. C., (2007). Glutamine, gene expression, and cell function. Bio Sci., 12: 344-357.

Curthoys, N. P., and Watford, M., (1995).Regulation of glutaminase activity and glutamine metabolism.Annu. Rev. Nutr., 15:133.

Cynober, L. A. (1999). The use of a-ketoglutarate salts in clinical nutrition and metabolic care. Curr. Opin. Clin. Nutr. Metab., Care 2: 33–38.

Dai, S., Gao, F., Zhang, W., Song, S., Xu, X. and Zhou, G., (2011). Effects of dietary glutamine and gamma-aminobutyric acid on performance, carcass characteristics and serum parameters in broilers under circular heat stress. Anim. Feed Sci. Tech., 168: 51-60.

Dai, S., Gao, F., Xu, X., Zhang, W., Song, S., Zhou, G., (2012). Effects of dietary glutamine and gamma-aminobutyric acid on meat colour, pH, composition, and water-holding characteristic in broilers under cyclic heat stress. Bri. Poult. Sci., 53:471-481.

Dai, S., Wang, L., Wen, A., Wang, L., Jin, G., (2009). Dietary glutamine supplementation improves growth performance, meat quality and colour stability of broilers under heat stress. Bri.Poult. Sci., 50:333-340.

Devi-Priya, K., Selvaraj, P., Nanjappan, K., Jayachandran, S., Visha, P., (2010). Oral supplementation of putrescine and L-glutamine on the growth performance, Immunity, intestinal enzymes in the broiler chickens. Tamilnadu J. Vet. Anim. Sci., 6: 250-254.

Dı´az-Acosta, A., Sandoval, M. L., Delgado-Olivares, L., and Membrillo-Herna´ndez, J., (2006). Effect of anaerobic and stationary phase growth conditions on the heat shock and oxidative stress responses in Escherichia coli K-12. Arch. Microbiol., 185:429–438.

Dirain, C. P. O., and Waldroup, P. W., (2002). Protein and amino acid needs of broiler in warm weather: A review. Inter. J. Poult. Sci., 4: 40-46.

Dokladny, K., Moseley, P.L., and Ma, T. Y., (2006). Physiologically relevant increase in temperature causes an increase in intestinal epithelial tight junction permeability. Am. J Physiol. Gastrointest. Liver Physiol., 290: 204–212.

Donkoh, A., (1989). Ambient temperature: a factor affecting performance and physiological response of broiler chickens. Intl. J. Biomet., 33: 259-265.

Eagle, H., Oyama, V .I., Levy, M., Horton, C .L., Fleischman, R., (1956). The growth response of mammalian cells in tissue culture to L-glutamine and L-glutamic acid. J. Biol. Chem., 218: 607.

Ebadiasl, G., (2011). Effects of supplemental glutamine and glutamate on growth performance, gastrointestinal development, jejunum morphology and Clostridium perfringens count in caecum of broilers. Degree Diss., Swedish University of Agricultural Science, Uppsala, Sweeden.

Edelman, I., and Culbertson, M. R., (1991). Exceptional codon recognition by the glutamine tRNAs in Saccharomyces cerevisiae. EMBO J., 10: 1481–1491.

Edens, F. W., (1978). Adrenal insufficiency in young chicken exposed to a high ambient temperature. Poult. Sci., 57: 17746-1750.

Ehrenfried, J. A., Chen, J., Li J., Mark Evers, B., (1995). "Glutamine-mediated regulation of heat shock protein expression in intestinal cells." Surg., 118: 352-357.

Ehrensvard G, Fischer A, Stjernholm R., (1949). Protein metabolism of tissue cells in vitro. The chemical nature of some obligate factors of tissue cell nutrition.Acta. Physiol. Scand., 18: 218–230.

Etches, R.J., John, I .M., and Gibbins, A. M. V., (1995). Behavioural, physiological, neuroendocrine and molecular responses to heat stress. In: N.J. Daghir (eds), Poultry production in hot climates, (CAB International Wallingford, UK), 31–65.

Fernandes, JIM., Scapini,L.B., Gottardo,E.T., Burin, A.M., Junior, Marques, F.E and Gruchouskei, L., (2013). Thermal conditioning during the first week on performance, heart morphology and carcass yield of broilers submitted to heat stress. Acta Sci. Anim. Sci., 35:311-319.

Fischer da Silva, A., Borges, S., Maiorka, A., Givisiez, P., Rocha, C., and Macari, M., (2007). Ornithine decarboxylase expression in the small intestine of broilers submitted to feed restriction and glutamine supplementation. Brazil. J. Poult. Sci., 9:111-115.

Formela, L. J., Galloway, S. W., and Kingsnorth, A. N., (1995).Inflammatory mediators in acute pancreatitis. Br. J. Surg., 82:6-13.

Furlan, R. L., (2006). Influencia da temperature na producao de frangos de corte. In Ananis 7 Simposio Brasil Sul de Avicultura, Empresa Brasileira de Pesquisa Agropecuaria (Embrapa/CNPSA), Chapeco, Brasil (CD-ROM).

Furlan, R. L., Carvalho, N. C., Malheiros, E. B. and Macari, M., (2001). Efeito da restricao alimentar inicial e da temperature ambiente sobre o desenvolvimento de visceras e ganho compensatorio em frangos de corte.Arq.Bras. Med. Vet. Zoot., 53:1-7.

Gebhardt, R. and Mecke, D., (1983). Heterogeneous distribution of glutamine synthetase among rat liver parenchymal cells in situ and in primary culture. EMBO J., 2: 567–570.

Geraert, P. A., Guillamin, S., and Leclercq, B., (1993). Are generically lean broilers more resistant to hot climate? Br. Poult. Sci. 34:643-653

Glover, J.R., and Lindquist S., (1998). Hsp104, Hsp70, and Hsp40: a novel chaperone system that rescues previously aggregated proteins Cel., 94: 73–82

González, F.H., Tecles, F., Martínez-Subiela, S., Tvarijonaviciute, A.., Soler, L., and Cerón, J. J., (2008).Acute phase protein response in goats.J. Vet. Diagno.Investi., 20: 580-584.

Gross, W. and Siegel, H., (1983). "Evaluation of the heterophil / lymphocyte ratio as a measure of stress in chickens."Avi. Dis., 972-979.

Gruys, E., and Snel, F. W. J. J.,(1994).Animal models for reactive amyloidosis.Baillière's Clin .Rheumatol., 8: 599-611.

Gruys, E., Toussaint, M., Niewold, T. and Koopmans, S., (2005). Review: Acute phase reaction and acute phase proteins. J. Zhejiang Univer. Sci., B., 6: 1045.

Hall, D. M., Buettner GR, Oberley, L. W., Xu, L., Matthes, R. D., Gisolfi C. V., (2001).Mechanisms of circulatory and intestinal barrier dysfunction during whole body hyperthermia.Am J Physiol Heart Circ Physiol., 280: 509–521

Hamiel, C. R., Pinto, S., Hau, A., and Wischmeyer, P. E., (2009). Glutamine enhances heat shock protein 70 expression via increased hexosamine biosynthetic pathway activity. Am. J. Physiol. Cell Physiol., 297: 1509–1519.

Harrison, J. F. and Kennedy, M. J., (1994). In vivo studies of the acid–base physiology of grasshoppers: the effect of feeding state on acid–base and nitrogen excretion. Physiol. Zool., 67: 120–141.

Hartl, F.U., and Hayer-Hartl, M., (2002). Molecular chaperones in the cytosol: From nascent chain to folded protein. Sci., 295: 1852-1858.

Haussinger, D., Gerok, W., (1983). Hepatocyte heterogeneity in glutamate uptake by isolated perfused rat liver. Eur. J. Biochem., 136: 421–425.

Haussinger, D., Stoll, B., Stehle T., Gerok, W., (1989). Hepatocyte heterogeneity in glutamate metabolism and bidirectional transport in perfused rat liver. Eur. J. Biochem., 185: 189–195.

Häussinger, D., (1990). Nitrogen metabolism in liver: structural and functional organization andphysiological relevance. Biochem. J., 267: 281-290.

Hernández, F., López, M., Martínez, S., Megías, M.D., Catalá, P., Madrid, J., (2012).Effect of low-protein diets and single sex on production performance, plasma metabolites, digestibility, and nitrogen excretion in 1-to 48-day-old broilers.Poult. Sci., 91: 683-692.

Hill, J. A., (1983). Indicators of stress in poultry World's Poult. Sci. J., 39: 24-31.

Hinnebusch, B. F., Ma, Q., Henderson, J. W., Siddique, A., Archer, S. Y., and Hodin, R. A., (2002). Enterocyte response to ischemia is dependent on differentiation state. J. Gastro. Surg., 6:403–409.

Hirakawa, T., Rokutan, K., Nikawa, T., and Kishi, K., (1996). Geranylgeranylacetone induces heat shock proteins in cultured guinea pig gastric mucosal cells and rat gastric mucosa. Gastro, 111: 345-357.

Horowitz, M., (2003).Matching the heart to heat-induced circulatory load: heat-acclimatory responses.News in Physio. Sci., 18:215– 221.

Huff, G. R., Huff, W. E., Balog, J. M., Rath, N. C., Anthony, N. B., and Nestor, K. E., (2005).Stress response differences and disease susceptibility reflected by heterophil to lymphocyte ratio in turkeys selected for increased body weight. Poult. Sci., 84:709-717.

Ihara, T., Tsujikawa, T., Fujiyama, Y., and Bamba, T., (2000).Regulation of PepT1 peptide transporter expression in the rat small intestine under malnourished conditions. Digest, 61: 59–67.

Inoue, Y., Grant, J. P., and Snyder, P. J., (1993). Effect of glutamine-supplemented intravenous nutrition on survival after Escherichia coli-induced peritonitis. JPen- Parenter. Enter. 17: 41-46.

Jang, H., Kwak, J. H., Cho, E.Y., We, Y.M., Lee, Y.H., Kim, S.C., and Han, D.J., (2008). Glutamine induces heat-shock protein-70 and glutathione expression and attenuates ischemic damage in rat islets. Transpl.Proceed, 40: 2581-2584.

Jazideh, F., Farhoomand, P., Daneshyar, M., Najafi, G., (2014). The effects of dietary glutamine supplementation on growth performance and intestinal morphology of broiler chickens reared under hot conditions. Turkish J.Vet. Anim. Sci., 38: 264-270.

Jungermann, K., Katz, N., (1989). Functional specialization of different hepatocyte populations. Physiol. Rev., 69:708-764.

Karaca, A. G.I., Parker, H. M., Yeatman, J. B., and McDaniel, C. D., (2002). The effects of heat stress and sperm quality classification on broiler breeder male fertility and semen ion concentrations.Bri Poult Sci., 43:621-628.

Keshavarz, K., (1990). Managing in hot weather.Broiler industry. Pp: 24-32.

Khan, J., Linoshi, Y., Cui, L., Sando, K., Tagaki, Y., and Okada, A., (1999). Ananyl-glutamine-supplemented parenteral nutrition increases luminal mucus gel and decreases permeability in the rat small intestine. JPen. Parenter. Enter, 23: 24-31.

Kiang, J. G., and Tsokos, G. C., (1998). Heat shock protein 70 kDa: molecular biology, biochemistry, and physiology. Pharmacol. Ther, 80: 183 – 201.

Kitt, J., Miller, P. S., Lewis A., and Fischer R. L., (2002).Effect of glutamine on growth performance and small intestine villus height in weanling pigs.Pages 29-32 in Nebraska Swine Rep. Univ. Nebraska, London.

Krebs, H. A., (1935). Metabolism of amino-acids: The synthesis of glutamine from glutamic acid and ammonia, and the enzymic hydrolysis of glutamine in animal tissues. Biochem J. 29: 1951–1969.

Kregel, K.C., (2002). Heat shock proteins: Modifying factors in physiological stress responses and acquired thermotolerance. J. Appl. Physio., 92: 2177–2186.

Lambert, G., (2009).Stress-induced gastrointestinal barrier dysfunction and its inflammatory effects. J. Anim. Sci., 87:101-108.

Lara L. J., and Rostagno, M. H., (2013). Impact of Heat Stress on Poultry Production. Anim., 3: 356-369.

Larson, S. D., Li, J., Chung, D. H., and Evers, B. M., (2007).Molecular mechanisms contributing to glutamine-mediated intestinal cell survival. Amer. J.Physio-Gastro. Liv. Physio., 293:1262-1271.

Lavoinne, A., Meisse, D., Quillard, M., Husson, A., Renouf, S., and Yassad, A., (1998). Glutamine and regulation of gene expression in rat hepatocytes: the role of cell swelling. Biochem., 80:807–811.

Lazarus, R. S., and Launier, R., (1978). Stress-related transactions between person and environment.In perspectives in interactional psychology, ed. L.A. Pervin, M. Lewis, 287-327. New York. Plenium.

Li, P., Yin, Y. L., Li, D. F., Kim, S. W., and Wu, G., (2007).Amino acids and immune function. Br. J. Nutr. 98: 237-252.

Li, Q., Zhang, Q., Wang, C., Liu, X., Qu, L., Gu, L., Li, N., and Li, J., (2009). Altered distribution of tight junction proteins after intestinal ischaemia/reperfusion injury in rats. J. Cell Mol. Med., 13:4061-76.

Liew, P., Zulkifli, I., Omar, D. A., and Hair-Bejo, A .R. (2003). "Effects of early age feed restriction and heat conditioning on heat shock protein 70 expression, resistance to infectious bursal disease, and growth in male broiler chickens subjected to heat stress." Poult. Sci., 82: 1879-1885.

Linda C. Y., Jin-Town W., Shu-Chen W., and Yen-Hsuan N., (2012). Host-microbial interactions and regulation of intestinal epithelial barrier function: From physiology to pathology. World J Gastrointest Pathophysiol. 3: 27–43.

Linke, R. P., Bock, V., Valet, G., Rothe, G., (1991). Inhibition of the oxidative burst response of N-formyl peptide- stimulated neutrophils by serum amyloid-A protein. Biochem.Biophys. Res. Commun., 176:1100-1105.

Lin, Y., and Zhou, X.Q., (2006). Dietary glutamine supplementation improves structure and function of intestine of juvenile Jian carp (Cyprinus carpio var. Jian). Aquacul., 256:389–394.

Lin, H., Jiao, H., Buyse, J., and Decuypere, E., (2006). Strategies for preventing heat stress in poultry. World's Poult. Sci. J., 62: 71-86.

Mancini, G., Carbonara, A.O., and Heremans, J. F., (1965).Immunochemical quantitation of antigens by single radial immune diffusion. Immuno-chem., 2: 235-254.

Marchini, C. F. P., Silva, P. L., Nascimoto, M. R. B. M., Beletti, M. E., Silva, N. M., and Guimaraes, E. C., (2011). Body weight, intestinal morphology and cell proliferation of broiler chickens submitted to cyclic heat stress. Intl. J. Poult. Sci., 10: 455 – 460.

Marissa, N. R., Yusheng, J. B., and Kenneth R. D., (2005).Thermally induced injury and heat-shock protein expression in cells and tissues. Ann. N.y. acad. Sci., 1066: 222–242.

Mashaly, M. M., Hendricks, G. L., III., Kalama, M. A., Gehad, A. E., Abbas, A. O., Patterson, P.H., (2004). Effect of heat stress on production parameters and immune responses of commercial laying hens.Poult. Sci., 83, 889–894.

Matteri R. L., Carroll J. A., and Dyer C. J., (2000).Neuroendocrine responses to stress. In: Moberg GP, Mench JA, editors. The biology of animal stress. CABI Publishing, p. 43–76.

Matthews J.C., and Anderson K. J., (2002). Recent advances in amino acid transporters and excitatory amino acid receptors. Curr. Opin. Clin. Nutr. Metab. Care., 5: 77–84.

Maiorka, A., Silva, A. V. F., Santin, E., Borges, S. A., Boleli, I. C., Macari, M., (2000). Influencia da suplementac¸ao de glutamina sobre o desempenho e o desenvolvimento de vilos e criptasdo intestino delgado de frangos. Arq.Bras. Med. Vet. Zoot, 52:487–490.

Maxwell, M. H., (1993). Avian blood leucocyte responses to stress.World's Poult. Sci. J., 49:34–43.

Mayer, M. P. and Bukau, B., (2005). Review Hsp70 chaperones: Cellular functions and molecular mechanism. Cell.Molecu. Life Sci., 62: 670–684.

McDaniel, C. D., Hood, J. E., and Parker, H. M., (2004).An Attempt at Alleviating Heat Stress Infertility in Male Broiler Breeder Chickens with Dietary Ascorbic Acid. Intl J. Poult. Sci., 3: 593-602.

McEwen, B. S., (2000). The neurobiology of stress: From serendipity to clinical relevance. Brain Res., 886:172–189.

Medina, M. A., (2001). Glutamine and cancer. J. Nutr., 131: 2539-2542

Meddings, J. B., and Swain, M., (2000). Environmental stress-induced gastrointestinal permeability is mediated by endogenous glucocorticoids in the rat. Gastroenter., 119:1019-1028.

Meisse, D., Claeyssens, S., Husson, A. and Lavoinne, A., (1999). Glutamine, a regulator of acute phase protein synthesis. Clinic. Nutr., 18:111-112.

Meltzer, A., (1983). Thermoneutral zone and resting metabolic rate of broilers. Brit. Poult. Sci., 24: 471-476.

Menconi, A., Kallapura, g., Hernandez-Velasco, X., Latorre, J., Morgan, M., Pumford, N. R., Layton, S., Urbano, T., Caseres. M., Pixley. C., Barton. J., M Hargis. B., and Tellez, G., (2013). Effect of Glutamine Supplementation Associated with Probiotics on Salmonella Typhimurium and Nitric Oxide or Glutamine with Perinatal Supplement on Growth Performance and Intestinal Morphology in Broiler Chickens .Clin.Microbial. 2:2327-5073

Mitchell, M. A., and Carlisle, A. J., (1992).The effect of chronic exposure to elevated environmental temperature on intestinal morphology and nutrient absorption in

the domestic fowl (Gullus domesticus). Comp. Biochem. Physio. Part A: Molecu. Integr. Physio., 101: 137-142.

Moberg, G. P., (2000). Biological response to stress: implications for animal welfare. In: Moberg GP, Mench J.A, editors. The biology of animal stress. CABI Publishing. p. 123–46.

Moorman, A.F.M., de Boer, P.A.J., Watford, M., Dingemanse, M. A. & Lamers, W. H., (1994). Hepatic glutaminase mRNA is confined to part of the urea cycle domain in the adult rodent liver lobule. FEBS. Lett., 356: 76–80

Mostl, E., and Palme, R., (2002). Hoemones as indicators of stress.Domest. Anim. Endocrin., 23: 67-74.

Morimoto, R. I., Tissieres, A., and Georgopoulos, C., (1994). Progress and perspectives on the biology of heat shock proteins and molecular chaperones. In: The Biology of Heat Shock Proteins and Molecular Chaperones, edited by Morimoto RI, Tissieres A, and Georgopoulos C. Cold Spring Harbor, NY: Cold Spring Harbor Laboratory Press.

Morini, S., Yacoub, W., Rastellini. C., (2000). Intestinal microvascular patterns during hemorrhagic shock. Digesti.Diseas. Sci., 45:710–722.

Morrison, A.L., Dinges, M., Singleton, K.D., Odoms, K., Wong, H.R. and Wischmeyer, P.E., (2006). Glutamine's protection against cellular injury is dependent on heat shock factor-1. American Journal of Physiology: Cel. Physio., 290:1625–1632.

Mosser, D. and Morimoto, R.I., (2004).Molecular chaperones and the stress of oncogenesis. Oncog., 23: 2907–2918.

Musch, M. W, Hayden, D., Sugi, K., Straus, D., and Chang, E. B., (1998).Cell-specific induction of hsp72-mediated protection by glutamine against oxidant injury in IEC18 cells. Proc. Assoc. Am. Physic., 110: 136-139.

Murakami, A., Sakamoto, M., Natali, M., Souza, L. and Franco, J., (2007). Supplementation of glutamine and vitamin E on the morphometry of the intestinal mucosa in broiler chickens. Poult. Sci., 86: 488-495.

Murakami, M., Tomita, Y., and Cathcart, M. K., (1988). Human hybridoma suppressor factor (HSF) inhibits IL2 production in addition to suppressing immunoglobulin production. Hybrid., 7:595-608.

Murata, H., (2007). Stress and acute phase protein response: an inconspicous but essential linkage. Vet. J., 173: 473-474

Murata, H., N. Shimada, Yoshinoka, M., (2004). "Current research on acute phase proteins in veterinary diagnosis: an overview." Vet. J., 168: 28-40.

Murata, H., and Miyamoto, T., (1993). Bovine haptoglobin as a possible immunomodulator in the sera of transported calves. Br. Vet. J., 149: 277-283.

Nakai A., and Morimoto, R. I., (1983). Characterization of a novel chicken heat shock transcription factor, heat shock factor 3, suggests a new regulatory pathway. Mol. Cell. Biol., 13: 1983-1997.

Naka, N., Ohsawa, M., Tomita, Y., Kanno, H., Uchida, A., Myoui, A. and Aozasa, K., (1996). Prognostic factors in angiosarcoma: a multivariate analysis of 55 cases. J. Surg. Onco., 61: 170-176.

Newsholme, P., (2001). Why is L-glutamine metabolism important to cells of the immune system in health, postinjury, surgery or infection? J. Nutri., 131: 2515-2522.

Newsholme, P., Procopio, J., Ramos Lima, M. M., Pithon-Curi, T. C., and Curi R., (2003).Glutamine and glutamate – their central role in cell metabolism and function.Cel.Biochem. Funct, 21: 1–9.

Nissim, I., M. Hardy, pleasure J., Nissim, I., (1993). "Effect of glutamine on heat-shock-induced mRNA and stress proteins." J. Cellul. Physio., 157: 313-318.

Nitsan, Z., Benae • Avraham, G., Zoref, Z., Nir, I., (1991). Growth and development of the digestive organs and some enzymes in broiler chicks after hatching.Brit.Poult. Sci., 32:515-523.

National Research Council-NRC., (1994).Nutrient requirements of poultry.National Academy Press, Washington, D.C.

Oliveira, G.P., Dias, C.M., Pelosi, P., Rocco, P. R.,(2010). Understanding the mechanisms of glutamine action incritically ill patients. Ann. Acad. Bras. Cienc., 82: 417-430.

Ooue, A., Kuwahara, T. I., Shamsuddin, A. K. M., Inoue, Y., Nishiyasu, T., Koga, S., Kondo, N., (2007). Changes in blood flow in a conduit artery and superficial vein of the upper arm during passive heating in humans. Euro. J. Appli. Physio., 101:97–103.

Osman, A. M., Tanios, N. I., (1983).The effect of heat on the intestinal and pancreatic levels of amylase and maltase of laying hens and broilers.Comparative Biochemistry and Physiology Part A: Physio., 75:563-567

O'Reilly, E.L. and Eckersall, P.D., (2014). Acute phase proteins: A review of their function, behavior and measurement in chickens. Worl.Poult. Sci. J., 70: 27-44.

Pelham, H. R. B., (1986). Speculation on the functions of the major heat shock and glucose-regulated proteins. Cel., 46: 959–961

Petersen, H. H., Nielsen, J. P., and Heegaard, P. M., (2004).Application of acute phase protein measurements in veterinary clinical chemistry. Veteri. Resear., 35:163-187, ISSN 1746-6148.

Pineiro, M., Pineiro, C., Carpintero, R., Morales, J., Campbell, F. M., Eckersall, P. D., Toussaint, M, J., and Lampreave, F., (2007).Characterisation of the pig acute phase protein response to road transport. Vet. J., 173:669-674.

Pirkkala, L., Nykänen, P., and Sistonen, l., (2001). Roles of the heat shock transcription factors in regulation of the heat shock response and beyond. FASEB J., 15: 1118-1131.

Quinteiro-Filho, W., Ribeiro, A., Ferraz-de-Paula, V., Pinheiro, M., Sakai, M., Sá, L., Ferreira, A. and Palermo-Neto, J., (2010). Heat stress impairs performance parameters, induces intestinal injury, and decreases macrophage activity in broiler chickens. Poult. Sci., 89: 1905-1914.

Randall, L. L., and Hardy, S. J. S., (1986). Correlations of competence for export with lack of tertiary structure of the mature species: a study in vivo of maltose-binding protein in E coli. Cel., 46: 921–928.

Rath, N. C., Anthony, N. B., Kannan, L., Huff, W.E., Huff, G. R., Chapman, H. D., Erf, G. F. and Wakenell, P., (2009). Serum ovotransferrin as a biomarker of inflammatory diseases in chickens.Poult. Sci., 88: 2069–2074.

Reeds, P.J., Burrin, D.G. and Stoll, B., (1997). Enteral glutamate is the preferential source for mucosa glutathione synthensis in fed piglets. The Ame J. Physio., 273: 408-415.

Sapolsky, R., (2005). "Biology and Human Behavior: The Neurological Origins of Individuality, 2nd edition". The Teaching Company. "See pages 19 and 20 of Guide Book"

Romero, L.M. and J. M. Reed, (2005). Collecting baseline corticosterone samples inthe field: is under 3 min good enough? Comp. Biochem. Physio. A., 140: 73-79.

Roth, E., (2008). Non- nutritive effect of glutamine. J. Nutri., 138: 2025S-2031S.

Saco, Y., Docampo, M. J., fagrega, E., Manteca, X., Diestre, A., Lampreave, F., and Bassols, A., (2003).Effect of transport stress on serum haptoglobin and Pig-MAP in pigs. Anim. Welf., 12:403-409.

Sakamoto, M. I., Faria, D. E., Nakagi, V. S., Murakami, A. E., (2014). Sources of Trophic Action on Performance and Intestinal Morphometry of Broiler Chickens Vaccinated against Coccidiosis. Braz. J. Poult. Sci. 16: 389-396.

Sakamoto, M. I., Faria, D.E., Nakagi, V.S., Negrão, J.A., Araújo, R.B., Souza, K.M.R., Previero, T. C., (2011). Utilization of glutamine, associated with glutamic acid, on development and enzymatic activity in broiler chickens. Arq.Bras. Med. Vet. Zootec., 63: 0102-0935.

Sakamoto, M., Murakami, A., Silveira, T., Fernandes, J., Oliveira, C. d., (2006). Influence of glutamine and vitamin E on the performance and the immune responses of broiler chickens. Revista Brasileira de Ciancia Avacola 8:243-249.

SAS.Institute, (2004). SAS/STAT user's Guide. Version 9.2 for window (SAS instr. Inc., Cary, NC).

Secchi, A., Ortanderl, J. M., Schmidt, W., Gebhard, M., Martin, E., Schmidt, H., (2000). Effect of endotoxemia on hepatic portal and sinusoidal blood flow in rats. J. Surg. Res., 89:26–30.

Selye, H., (1973). The evolution of the stress concept.Ame.Scient, 61: 692– 699.

Shakeri, M., Zulkifli, I., Soleimani, A.F., O'Reilly, E.L. Eckersall, P.D., Kumari, S., Aryani, A.A. and Abdullah, F.F., (2014). Response to dietary supplementation of L-glutamine and L-glutamate in broiler chickens reared at different stocking densities under the hot, humid tropical conditions. Poult. Sci., pii: PS3910.

Shini, S., Shini, A., Kaiser, P., (2010). Cytokine and chemokine gene expression profiles in heterophils from chickenstreated with corticosterone. Stress 13: 185-194.

Shizuka, F., Vasupongsotorn, S., Kido, Y., Kishi, K., (1990).Comparative effect of intravenously or intragastrically administered glutamine on small intestinal function of the rat.The Tokushima J. Experim. Med., 37:49-57.

Selye, H., (1950). The physiology and pathology of exposure to stress. Oxford, England: Acta, Inc. xx 822: 203 pp.

Selye, H., (1973). The evolution of the stress concept. American Scientist, 61, 692–699.

Siegel, H.S., (1995). Stress, strains and resistance. Bri.Poult. Sci., 36: 3-22.

Siegel, H.S., (1980).Physiological stress in birds.Bio. Sci., 30: 529-534.

Siegel, H. S., and Van Kampen, M., (1984).Energy relationships in growing chickens given daily injections of corticosterone. Br. Poult. Sci., 25:477–485

Sikora, A., and E., Grzesiuk, (2007)."Heat shock response in gastrointestinal tract."J. Physio. Pharmaco., 58: 43.

Singleton, K. D. and Wischmeyer, P. E., (2006). Oral glutamine enhances heat shock protein expression and improves survival following hyperthermia. Shock 25: 295-299.

Sklan, D., (2001). Development of the digestive tract in poultry.World.Poult. Sci. J., 57: 415-428.

Skrzypek, T., Valverde Piedra, J.L., Skrzypek, H., Wolinski, J., Kazimierczak, W., Szymanczyk, S., Pawlowska, M., Zabielski. R., (2005).Light and scanning electron microscopy evaluation of the postnatal small intestinal mucosa development in pigs. j physiol pharmacol. 56 suppl., 3:71-87.

Smith, R.J., (1990). Glutamine metabolism and its physiologic importance. JPen.-Parenter. Enter, 14: 40-44.

Soares, A. D., Costa, K. A., Wanner, S. P., Santos, R. G., Fernandes, S. O., Martins, F. S., Nicoli, J. R., Coimbra, C. C., Cardoso, V. N., (2014). Dietary glutamine prevents the loss of intestinal barrier function and attenuates the increase in core body temperature induced by acute heat exposure. Br J. Nutr., 112:1601–1610.

Soleimani, A.F., I. Zulkifli, A.R. Omar and A.R. Raha, (2011).Physiological response of three breeds to 327-335.Acute heat stress.Poult. Sci., 90: 1435-1440.

Soleimani, A.F., Zulkifli, I., Hair-Bejo M, Omar, A.R. and Raha, A.R., (2012a). The role of heat shock protein 70 in resistance to Salmonella enteritidis in broiler chickens subjected to neonatal feed restriction and thermal stress. Poult. Sci., 91: 340-345.

Soleimani, A.F., Zulkifli, I., Omar, A.R. and Raha, A.R., (2012b). The relationship between adrenocortical function and hsp70 expression in socially isolated Japanese quail. Comp. Biochem. Physio., 161: 140-144.

Soltan, M., (2009). Influence of dietary glutamine supplementation on growth performance, small intestinal morphology, immune response and some blood parameters of broiler chickens. Intl. J. Poult. Sci., 8: 60-68.

Souba, W. W., Klimberg, V. S., Hautamaki, R. D., Mendenhall, W. H., Bova, F. C., Howard, R. J., Bland, K. I.,Copeland, E. M., (1990). Oral glutamine reduces bacterial translocation following abdominal radiation. J. Surg. Res., 48:1-5.

Stintzi, A., Marlow, D., Palyada, K., (2005). Use of genome-wide expression profiling and mutagenesis to study the intestinal lifestyle of Campylobacter jejuni. Infect. Immun., 73: 1797-1810

Sutisa, K., Supattra, O., Laddawan, H., Buntita, T., and Wittawat, M., (2011). Influence of supplemental glutamine on nutrient digestibility and utilization, small intestinal morphology and gastrointestinal tract and immune organ developments of broiler chickens. World Acad. Sci. Engineeri.Techno., 5:08-29.

Tapiero, H., Mathe, G., Couvreur, P. and Tew, K., (2002). II. Glutamine and glutamate. Biomed. Pharmacothe., 56: 446-457.

Teeter, R.G. and Belay, T. (1996). 'Broiler management during acute heat stress'. Anim. F. Sci. Tech., 58: 127-142.

Touchette, K. J., Carroll., J. A., Allee, G. L., Matteri, R. L., Dyer, C. J., Beausang, L. A., Zannelli, M. E., (2002). Effect of spray-dried plasma and lipopolysaccharide exposure on weaned pigs: I. Effects on the immune axis of weaned pigs. J. Anim. Sci., 80:494-501.

Towbin, H. Stachelin, T. Gordon, J. (1979). Electrophoretic transfer of proteins from polyacrilamide gels into nitrocellulose sheets: Procedure and some applications. Proc. Natl. Acad. Sci., 76:4350–4354.

Tîrziu, E., (2009). Acute phase proteins in immune response.Lucrări tiinłifice medicină veterinară vol. Xlii (1), timioara. 329.

Turner, J. R., (2009). Intestinal mucosal barrier function in health and disease.Natur. Rev. Immuno., 9: 799-809.

Uni, Z., (1999). Functional development of the small intestine in domestic birds: cellular and molecular aspects. Poult. Av. Biolo. Rev., 10:167-179.

Uni, Z., Noy, Y., and Sklan, D., (1995). Posthatch Changes in Morphology and Function of the Small Intestines in Heavy- and Light-Strain Chicks.Poult. Sci., 74: 1622-1629

Uni, z., Platin, R., Sklan, D., (1998). Cell proliferation in chick intestinal epithelium occurs both in crypts and along the villus. J. Comp. Physiol., 168:241.

Upragarin N, Toussaint MJM, Tooten, P.C.J., Lamdman, W. J., Gruys, E., (2005). Acute Phase Protein Reaction in Layer Chickens. A Calculated Acute Phase Protein Index as Measure to Assess Health during the Rearing Period; Proceedings 14th World Veterinary Poultry Congress; Istabul, Turkey.. p. 344.

Vabulas, R.M., Raychaudhuri, S., Hayer-Hartl, M. and Hartl, F.U., (2010). Protein folding in the cytoplasm and the heat shock response.Cold Spring Harb.Perspect. Biol., 2: a004390.

Verlander J. W., Chu, D., Lee, H., Handlogten, M. E., and David Weiner, I. D., (2013). Expression of glutamine synthetase in the mouse kidney: localization in multiple epithelial cell types and differential regulation by hypokalemia. Am J Physiol Renal Physiol. 5: 701–713.

Virden, W. S. and Kids, M. T., (2009). Physiological stress in broilers: Ramifications on nutrient digestibility and responses. J. Appl. Poult. Res., 18: 338–347.

Wang, S., Bottje, W., Maynard, P., Dibner, J., Shermer, W., (1997).Effect of santoquin and oxidized faton liver and intestinal glutathione in broiler.Poult. Sci., 76:961-967.

Wang, J.J., Chen, L.X., Li, P., Li, X.L., Zhou, H.J., Wang, F.L., Li, D.F., Yin, Y.L., Wu, G., (2008). Gene expression is altered in piglet small intestine by weaning and dietary glutamine supplementation. J. Nutr., 138:1025-1032

Wang, W. W., Qiao, S. Y., Li, D. F., (2009). Amino acids and gut function. Amino Acid, 37:105–110.

Watford, M., (2008). Glutamine metabolism and function in relation to propline synthesis and the safety of glutamine and proline supplementation. J. Nutr., 138: 2003-2007.

Wilmore, M. D., Douglas, W., Judith, K., Shabert, M. D., and Mph, R. D., (1998). Role of glutamine in immunologic responses. Nutr, 14: 7-8.

Windisch, W., Schedle, K., Plitzner C., and Kroismayr, A., (2007).Use of phytogenic products as feed additives for swine and poultry. J. Anim. Sci. 86:140–148

Wischmeyer, P.E., (2002). Glutamine and heat shock protein expression. Nutriti., 18: 225-228.

Wischmeyer, P.E., Kahana, M., Wolfson, R., Ren, H., Musch, M. M. and Chang, E.B., (2001). Glutamine induces heat shock protein and protects against endotoxin shock in the rat. J. Appl. Physio., 90: 2403-10

Wright, P. A., (1995). A review. Nitrogen excretion: Three end products, many physiological roles. J. of Experiment. Bio., 198: 273 - 281

Wu, G. (1998). "Intestinal mucosal amino acid catabolism." J. Nutr., 128: 1249-1252.

Wu G. and Morris, Jr. S. M., (1998).Arginine metabolism: nitric oxide and beyond.Biochem. J., 336, 1-17.

Wu, G., Bazer, F. W., Davis T. A., Jaeger, L A., Johnson G. A., Kim, S.W., Knabe, D. A., Meininger C J., Spencer, T. E., Yin Y., (2007). Important roles for the arginine family of amino acids in swine nutrition and production.Livestock. Sci., 112: 8-22.

Wu, G., Fang, Y, Z., Yang, S., Lupton, J. R., Turner, N, D., (2004). Gluthathione metabolism and its implications for health. J. Nutr., 134:489-492.

Wu, G., Meier, S. A., Knabe. D. A., (1996). Dietary glutamine supplementation prevents jejunal atrophy in weaned pigs. J. Nutr., 126:2578-2584.

Wu, X., Xie, C., Zhang, Y., Fan, Z., • Yin, Y., Blachier, F., (2015). Glutamate–glutamine cycle and exchange in the placenta–fetus unit during late pregnancy. Amin. Acid., 47:45-53.

Xie, H., Huff, G.R., Huff, W.E., Balog, J.M. and Rath, N.C., (2002).Effects of ovotransferrin on chicken macrophages and heterophil-granulocytes. Dev. Comp. Immuno., 26: 805–815.

Yahav, S., and McMurtry, J. P., (2001).Thermotolerance acquisition in broiler chickens by temperature conditioning early in life- the effect on timing and ambient temperature.Poult. Sci., 12: 1662-1666.

Yamauchi, K., (2002). Review on chicken intestinal villus histological alterations related with intestinal function. J. Pout. Sci., 39: 229-242.

Yi, G. F., Allee, G. L., Frank, J. W., Spencer, J. D., Touchette, K. J., (2001). Impact of glutamine, menhaden fish meal and spray-dried plasma on the growth and intestinal morphology of broilers.Poult. Sci., 80:201.

Yi, G., Carroll, J., Allee, G., Gaines, A., Kendall, D., Usry, J., Toride, Y. and Izuru, S., (2005). Effect of glutamine and spray-dried plasma on growth performance,

small intestinal morphology, and immune responses of Escherichia coli K88+-challenged weaned pigs. J. Anim. Sci., 83: 634-643.

Yu J., Bao E. D., Yan J. Y., Lei L. (2008). Expression and localization of Hsps in the heart and blood vessel of heat-stressed broilers. Cell Stres. Chaperon., 13:327–335.

Zuhl, M.N., Dokladny, Mermier CM, Schneider S, Salgado R, Moseley P. L., (2015). The effects of acute oral glutamine supplementation on exercise-induced gastrointestinal permeability and heat shock protein expression in peripheral blood mononuclear cells. Cell Stres. Chaperon., 20:85–93.

Zuhl, M.N., Lanphere, K.R., Kravitz, L., Mermier, C. M., Schneider, S., Dokladny, K., Moseley, P. L., (2014). Effects of oral glutamine supplementation on exercise-induced gastrointestinal permeability and tight junction protein expression.J. Appl. Physiol., 116:183–191.

Zulkifli, I., Al-Aqil, A., Omar, A. R., Sazili, A. Q., (2009). "Crating and heat stress influence blood parameters and heat shock protein 70 expression in broiler chickens showing short or long tonic immobility reactions." Poult. Sci. 88: 471-476.

Zulkifli, I., Che Norma, M.T., Israf, D.A., Omar, A.R., (2000). The effect of early age feed restriction on subsequent response to high environmental temperatures in female broiler chickens. Poult. Sci., 79: 1401–1407.

Zulkifli, I., Che Norma, M.T., Israf, D.A., Omar, A.R., (2002). The effect of early age feed restriction on heat shock protein 70 response in heat-stressed female broiler chickens. Br. Poult. Sci., 43: 141–145.

Zulkifli, I., Dunnington, E.A., Gross, W.B., and Siegel, P.B., (1994). Food restriction early or later in life and its effect on adaptability, disease resistance, animmunocompetence of heat stressed dwarf and nondwarf chickens.Br. Poult. Sci. 35: 203–214

Zulkifli, I., and Siegel, P. B., (1995). Is there a positive side to stress? World's Poult. Sci. J. 51:63–76.

Zulkifli, I., Liew, P. K., Israf, D. A., Omar A. R. and Hair-Bejo, M., (2003). Effects of early age feed restriction and thermal conditioning on heterophil/lymphocyte ratio, heat shock protein 70 and body temperature of male broiler chickens subjected to acute heat stress. J. Therm. Biol., 28: 217-222.

Zulkifli, I., Najafi, P., Nurfarahin, A.J., Soleimani, A.F., Kumari, S., Aryani, A.A., O'Reilly, E.L. and Eckersall, P.D., (2014). Acute phase proteins, interleukin 6, and heat shock protein 70 in broiler chickens administered with corticosterone. Poult. Sci., 93:3112-3118.

Zulkifli, I., Ramlah, A.H., (1998). Biological and economic performance of broiler chickens fed mashed or pelleted diet. Malays. Appl. Biol., 27:21-24.