Cairo University
Faculty of Veterinary Medicine
Dept. of Nutrition and Clinical Nutrition

"Effects of Guanidinoacetic acid supplementation on broiler chickens fed on all-vegetarian diets under normal and challenging conditions"

Thesis

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Abstract

A feeding trial was conducted to investigate the response of broiler chickens to dietary fortification of guanidinoacetic acid (GAA) in a commercial form CreAMINO® and highlighting its impact on growth, serum parameters, Total Creatine Kinase(CK), Cardiac Creatine Kinase (CK- MB) and triiodothyronine (T3)], oxidation stress biomarkers, mitochondrial functions, carcass characteristics, proximate beast muscles composition and the related histomorphological picture as well as histopathological changes of liver tissues at the end of 32 days experimental period under ideal (normal and challenging condition (induced by 1.5 mg/kg dietary fortification of (T3) hormone). A total of 192 one-day old sexed Ross 308 broiler chicks were randomly assigned into 4 equal groups each of 8 replicates (6 chicks/ replicate). The first (G1) served as the control and fed on basal diets without any supplementation, the second (G2) was fed on basal diets supplemented on top with (GAA) that was commercially available CreAMINO® product (600 g/Ton) and the third group (G3) was fed on same diets and fortified with 1.5 ppm triiodothyroinine (T3) as challenging agent. Whereas, the fourth (G4) was fed on the same diets and supplemented with both triiodothyronine and guanidinoacetic acid at the aforementioned levels.

Results showed that the overall final body weights and weight gains in GAA supplemented groups were tended to be improved as a result of such supplementation ($P = 0.05$), however feed intake was not significantly affected ($P > 0.05$), but the FCRs were significantly improved ($P < 0.0001$) as compared to non-supplemented. The addition of (T3) hormone resulted in an expected highly significant negative impact on all the examined growth parameters ($P < 0.0001$) as compared to non-challenged groups. A trend to partial improvement in weight gains and feed intake were noticed as a result of GAA fortification, but failed to overcome the adverse effects of (T3) hormone on the growth parameters in a complete and significant manner. The GAA fortified groups showed a significant ($P < 0.0001$) reduction in the levels of Cardiac Creatine Kinase (CK- MB) compared to the non-supplemented, a situation that confirmed the protective ability of such additive against the retrogressive changes of cardiac muscles. Meanwhile; GAA supplementation tended to reduce ($P=0.07$) the level of total CK. Meanwhile creatinine levels seemed to be non-significantly ($P > 0.05$) altered. Also GAA supplemented diets had no influence on AST, ALT, total protein and serum T3 hormone level ($P > 0.05$). However, such supplement showed a trend to reduce ALT level. Dietary T3 treatment significantly increased ALT ($P = 0.01$) and Total CK ($P < 0.0001$) as compared to untreated groups. Additionally T3 treatment increased serum T3 significantly ($P < 0.0001$). An interaction (GAA X T3) between groups receiving GAA and those treated with T3 concerning their effects on the levels of total CK, CK-MB were noticed. It was clear that GAA supplementation at such rate had the ability to reduce the level of total CK (G2) a situation that indicated its protective effect against degenerative changes in the body (mainly in cardiac and skeletal muscles as well as brain tissue) however; GAA together with T3 hormone treatment had no effect on total CK. On the other hand, the levels of cardiac CK (CK-MB) were significantly ($P < 0.0001$) reduced as a result of GAA supplementation either solely or in combination with T3 hormone. It seemed

that the GAA has the ability to protect heart muscle against degenerative changes in some way but failed to play the same protective role with skeletal muscles under the T3 hormone challenging condition. Regarding the impact on cardiac creatine kinase activity %(CK-MB %) the main effects of GAA dietary supplemented diets and T3 hormone treatment diets were significantly lower than un-supplemented groups (P < 0.0001 and P= 0.001) respectively. A significant interaction effects (GAA X T3 hormone) was noticed (P=0.002). Birds in (G4) showed the lowest value among groups significantly. These findings further confirmed the protective effect of GAA on cardiac muscle. GAA fortification showed a significant (P=0.003) increase in reGSH and a significant (P=0.02) lowest value of MDA however, both GPx and SOD showed only a trend to be improved (P=0.06 and P=0.09) respectively compared to non-supplemented, suggested the advantageous effect of GAA in modulating the anti-oxidant system especially under stress condition. T3 hormone has significant [(reGSH; P=0.004), (GPx; P=0.001); P=0.003)] effect on reGSH, GPx and MDA but, SOD was not affected. Interaction (GAA X T3) were only observed in MDA (P =0.03) and SOD (P=0.02).

A highly significant positive effect on [(Complex I; P< 0.0001), (complex II; (P <0.001), (complex III; P = 0.030), complex IV; P <0.001) and Mitochondria RCR; P< 0.0001). Also T3 had a severe suppressive effect (P<0.0001) on mitochondrial complexes and mitochondrial RCR. An interaction (GAA X T3 hormone) was noticed in [(complex I; P< 0.0001), (complex III; P<0.0001), (complex IV, P= 0.01) and (mitochondrial RCR; P=0.02)]. No interaction was observed in complex II (P> 0.05). The lowest activity was markedly noticed in G3. No mortalities, gross lesions and left ventricle abnormalities were recorded in (G2) that fortified with GAA Meanwhile, the survival rates of T3 treated groups were 47.92% and 46.83% respectively a condition that means the GAA fortification to diets treated concurrently with T3 hormone did not affected the mortality rates. In addition, most of dead birds as a result of T3 treatment showed RVH (right ventricular hypotrophy) and ascites starting from the second (2^{nd}) up to the fifth ($5^{th)}$ weeks of age. The GAA fortified groups showed a significant reduction in abdominal fat yield (P<0.0001) meanwhile; a significance increase in heart index (P=0.045) was noticed. The other carcass traits remained unaffected. Dietary T3treatment negatively affected the most of carcass traits significantly (P< 0.0001). An interaction was only observed in heart index (P=0.01). It was clear that the dietary GAA supplementation seems to be not affected the proximate composition of breast meat. Dietary GAA showed normal histological structure of liver tissue. Meanwhile liver samples of birds with dietary T3 treatment showed thickening of the bile ducts with collagen and oedema in hepatic capsule together with vaculation in the cytoplasm of some hepatocytes with other coagulative necrosis underneath the thick capsule. The combined impact of both GAA + T3 hormone fortification showed that dilatation in the central vein and portal vein as well as sinusoids with thickening in the vascular wall together with infiltration of red blood cells in the hepatic sinusoids.

It could be concluded that, GAA in a commercial form CreAMINO®, could be used in as an efficacious precursor for creatine in broiler chickens under normal and stressful conditions. It has the potential to induce a positive effects not only on growth performance, some selected serum parameters, carcass traits, carcass composition but also alleviating oxidative stress as indicated by improved antioxidant biomarkers and modulating the mitochondrial activity as well as partially ameliorated the pathological changes in the liver occurred after challenging with dietary T3 hormone fortification of the birds and proved to be economically feasible.

Key words: Antioxidant biomarkers, broiler, Carcass traits, CreAMINO®, Growth, Guanidinoacetic acid, histopathology, mitochondrial activity, T3 hormone

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