 doi:10.20944/preprints202105.0366.v1

Review

Potential effects of delphinidin-3-*O*-sambubioside and cyanidin-3-*O*-sambubioside of *Hibiscus sabdariffa* L. in ruminant meat and milk production and quality

Rosalba Lazalde-Cruz [1], Deli Nazmín Tirado-González [2,*], Germán David Mendoza-Martínez [3], Alejandro Lara-Bueno [4], María Isabel Carrillo-Díaz [5], Gustavo Tirado-Estrada [6,*], Luis Alberto Miranda-Romero [7,*]

1 Universidad Autónoma Chapingo- Posgrado en Producción Animal, Departamento de Zootecnia, Estado de México, México. C.P. 56320; e-mail: rlazaldc.909@gmail.com
2 Instituto Nacional de Investigaciones Forestales Agrícolas y Pecuarias (INIFAP)- CENID AF; C.P. 47540; e-mail: tirado.deli@inifap.gob.mx
3 Universidad Autónoma de México- Xochimilco, Cd. México, México. C.P. 04960; e-mail: gmendoza@correo.xoc.uam.mx
4 Universidad Autónoma Chapingo- Posgrado en Producción Animal, Departamento de Zootecnia, Estado de México, México. C.P. 56320; e-mail: alarab_11@hotmail.com
5 Universidad de Colima- Facultad de Medicina Veterinaria y Zootecnia, Colima, México; C.P. 28100; e-mail: mcarrillo13@ucol.mx
6 Tecnológico Nacional de México/Instituto Tecnológico El Llano Aguascalientes, Aguascalientes, México; C.P. 20330; e-mail: gustavo.te@llano.tecnm.mx
7 Universidad Autónoma Chapingo- Posgrado en Producción Animal, Departamento de Zootecnia, Estado de México, México. C.P. 56320; e-mail: microbiologia.pecuaria08@gmail.com

* **Correspondence** tirado.deli@inifap.gob.mx; deli_gym@hotmail.com (Tirado-González, D.N., Ph.D.); gustavo.te@llano.tecnm.mx (Tirado-Estrada, G., Ph.D.); microbiologia.pecuaria08@gmail.com (Miranda-Romero, L.A., Ph.D.).

Abstract: The objective was to analyze the effects of adding anthocyanin delphinidin-3-*O*-sambubioside and cyanidin-3-*O*-sambubioside of *Hibiscus sabdariffa* L. in animal diets. Scientific articles published before 2021 in clinics, pharmacology, nutrition, and animal production were included. The grains/concentrate, metabolic exigency, and caloric stress contribute to increasing the reactive oxygen species (ROS); the excess of ROS unbalance the oxidants and antioxidants. Cyanidin-3-*O*-sambubioside and delphinidin-3-*O*-sambubioside have antioxidant, antibacterial, antiviral, and anthelmintic activities. In the rumen, anthocyanin might show interactions and/or synergisms with substrates, microorganisms, and enzymes which could reduce the fiber degradability, but increase the potential methane (CH_4) emissions; since anthocyanin interferes in the biohydrogenation of fats, they increase the fat milk and meat quality. Anthocyanins reduce plasma oxidation and deposit in tissues, increasing the milk and meat antioxidant activities. Cyanidin-3-*O*-sambubioside and delphinidin-3-*O*-sambubioside act as inhibitors of the angiotensin-converting enzyme (ACEi) and rennin expression which may improve milk yield (there is not enough evidence in ruminants, though). Polyphenols affect the reproductive potential. Sub products of HS contain as many amounts of polyphenols as calyces, and their inclusion in diets would positively affect the average daily gain and fat meat quality. Including HS in ruminant diets can improve the meat and milk quality.

Keywords: *Hibiscus sabdariffa* L.; agricultural wastes; anthocyanins; ruminant nutrition; milk and meat production; fat milk and meat quality.

1. Introduction

Hibiscus sabdariffa L. (HS) is a type*Hibiscus sabdariffa* L. (HS) is a type of shrub of *Malvaceae* family from India [1, 2], adapted to spring-summer and subtropical or tropical environments (Aw/As (Köppen climate classification)) [2-4]. In Mexico, HS production has increased 10.54% from 2003 to 2018 [5, 6]. of shrub of *Malvaceae* family from India [1, 2], adapted to spring-summer and subtropical or tropical environments (Aw/As (Köppen

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 17 May 2021 doi:10.20944/preprints202105.0366.v1

climate classification)) [2-4]. In Mexico, HS production has increased 10.54% from 2003 to 2018 [5, 6].

According to FAO [7], the HS calyxes are one of the most demanded products by industry to human feeding [8, 9]. Due to fatty acids HS contents and proportions [10, 11], antioxidants [12-14] and antimicrobial (Gramm negative bacteria) [15], antiviral [16], and anthelmintic properties [17], which might have improvements on human wealth [12, 18, 19].

Calyxes of HS contain 15.76-0.04% of a-linoleic fatty acid (n-3) [10, 11], and flavonoids classified as anthocyanins [20]. Factors as the type of HS variety, crop management, processing, storage, extraction of extracts, and cell contents affect the antioxidant contents [21-23], however, the most proportion of HS flavonoids are the anthocyanins cyanidid-3-*O*-sambubioside (25.9 to 46.2%) and delphinidin-3-*O*-sambubioside (48.4 to 59.2%) [13, 23-25] whose clinic effects on humans are different from another kind of flavonoids supplements as green tea (*Camelia sinensis*) which mainly contain epigalocathequin-3-gallate (EGCG) and epicahequin-3-gallate [26].

Overall polyphenols and another kind of antioxidants as selenium and a-tocopherol reduce the free radicals and chelate pro-oxidant metals [9, 14, 27-29], can affect the ruminal digestibility and fermentation kinetics [30, 31], and in the animal productive behavior [32], reducing the effects of the oxidative stress in ruminants [33-35] caused by the high grain and concentrates proportions on diets [26, 36, 37], the metabolic exigency, and the heat stress [37], and therefore improving the oxide-reductive potential of products derived to human feed [38].

The objective of the present study was to make a critical review about the potential clinic and productive effects of including HS anthocyanin: cyanidin-3-*O*-sambubiosid and delphinidin-3-*O*-sambubiosid in ruminant diets.

2. Oxidative stress in ruminants

Inflammatory and environmental processes increase the endogenous ROS. Unbalance between pro-oxidants and antioxidants might promote oxidative stress and molecular damage [37].

In dairy cows and beef cattle, the environmental pollution and the high metabolic exigency during pregnancy, milk production, negative energy balance, heat stress, respiratory diseases, inflammatory process, and parasites promote ROS releasing (O_2, OH, RO_2, RO, HO_2, H_2O_2, HOCl, O_3, etc.), meanwhile the adipose mobilization increases the pro-inflammatory cytokines [37]. Potential negative effects on animal wealth would worse in the future because of the population increment and therefore the milk and meat demand [27].

ROS contribute to inflammatory processes through the necroptosis activation (NF-κβ) via phosphorylation interleukin (I-κβ), and because of the production of pro-inflammatory cytokines such tumoral factors (TNF-α). In addition, protein carbonylation is mediated by the ROS and metals (Fe^{2+}, Cu^{+}, etc.), producing oxidative by-products and advanced oxidative protein products (AOPP): 1) carbohydrates and lipids have reactive compounds to carbonyl from glycoxidation and lipoperoxidation that might bond to protein residues; 2) oxidized proteins are degraded by proteases, but chemically modified proteins (by di-tyrosine and disulfide cross linkages) might not be substrates to proteolysis, contributing to deposits in tissues and organ damages [39-44].

High grain and concentrates proportion in ruminant diets increases the lipoperoxidation, decreasing the α-tocopherol and the ferric reductive availability in blood plasma [22] and increasing the amount of AOPP, negatively related with milk yield because of the oxide-reductive unbalance. Including high-grain diets and therefore the reduction of forage proportion rises the abnormal amount and types of metabolites in rumen [37].

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 17 May 2021 doi:10.20944/preprints202105.0366.v1

In viral, bacterial, and fungal infections, phagocytes and neutrophils are sources of ROS that interfere in a chain of chemical reactions that increase the hypochlorous oxidant potential which might be useful to combat the photogenes, but damaging tissues. Besides this, parasites induce inflammatory followed by eosinophils increasing which also contribute to tissues damage. Lactation and heat stress are potential sources of AOPP, thereby of TNF-α expression and potential mammal glandule diseases, increasing the milk and meat contents of ROS [45, 46].

Oxidized milk and meat contribute to a higher ROS content in blood plasma which would be a threating to human welfare [38].

3. Potential clinic effects of antioxidants

Polyphenols are a wide variety of secondary plant metabolites with at least one -OH that can be structurally simple (egallic and gallic acids) or complex (dymers, olygomeric, and polymeric with high molecular weight). Antioxidants can be classified as flavonoids or non-flavonoids, thus flavonoids can be flavones, flavanones, isoflavones, flavonols, flavan-3-ols and anthocyanins (from flavan-3-ols derived the condensed tannins (non-hydrolysable)), phenolic acids, hydrolysable tannins, and stilbenes are clustered as non-flavonoids [17].

Because of the structural differences among complexes, total phenolic compounds cannot directly be related with total antioxidant availability [21, 22]. The EGCG, primarily found in green tea had a galloyl group in the third position and an o-trihydroxy in the β-ring which protect cell from ROS damage [42, 47]; by the regulation the over expression of genes EGCG have anti-inflammatory and antioxidant effects in reduction of apoptosis, cell fibrosis, and tumoral growing via regulation and reduction of kinases, signal transduction, and transcription activation [44, 48]. EGCG can [49]:

1) Promote the cytotoxicity to increase the antitumoral activities, by producing H2O2 with its pyrogallol moiety or the reduction of Fe(III) to Fe(II), generating -OH ROS (although cysteine N-acetyl protect cells from cytotoxicity of H2O2 it does not avoid cell death process).
2) Promote apoptosis through mitochondrial damage, membrane depolarization, and cytochrome c release, and protects against mitochondrial damage-related cell death without changes in SOD, glutathione peroxidase, Nrf2, Bcl2, and oxidative stress. Modulates gene expression by inhibiting various transcription factors (including Sp1, NF-κB, AP-1, STAT1, STAT3, and FOXO1) and the expression of NF-κB and AP-1. EGCG inhibits STAT1 to mediate protective effects on myocardial injury.
3) By increasing second messengers, such as Ca^{2+}, cAMP, and cGMP. EGCG elevates cytosolic Ca^{2+} without electrical stimulation by inhibition of SERCA (Ca^{2+}-ATPase activity), which affects the activities of Ca^{2+}-requiring enzymes, such as calmodulin (CAM)-dependent protein kinase II and CAMKKβ (CAMKKβ is an upstream regulator of AMP-dependent kinase (AMPK), which plays crucial roles in energy metabolism and cardiovascular functions). Stimulates vasorelaxation by increasing cAMP and cGMP in the aorta, then it may stimulate the production of cyclic nucleotides with beneficial biological effects in cardiovascular physiology.
4) Inhibit the transcription of FOXO1 to lead the suppression of basal levels of endothelin-1 and differentiation of adipocytes. In mitochondria, EGCG enhances fat utilization, reducing the expression of leptin and stearyl-CoA desaturase while increasing fat oxidation.
5) EGCG inhibits DNA methyltransferase, which reverses methylation-induced gene silencing.
6) Inhibits autophagy, leading to apoptosis in macrophage cell lines.

Although the extracts of HS also change the oxidative potential of blood plasma, increasing the glutathione intracellular, but its primarily action is on Renin-Angiotensin-

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 17 May 2021 doi:10.20944/preprints202105.0366.v1

Aldosterone System (RAS) interfering the electrolytic regulation, blood pressure, and the cardiac function [50], the increasing of adrenalin, catecholamines, and noradrenalin (by specific Angiotensin (AngII)) [51].

Guerrero et al. [52] tested the activity of the Angiotensin Converting Enzyme inhibitor (ACEi) of 17 different types of flavonoids, the ACEi increased when: 1) the catechol group was in the β-ring (3’, 4’-dihidroxy); 2) there is a doble bond between C2 and C3 of carbon rings; and 3) there is a ketone in the C4 of the carbon ring. The absence of C4 in the carbonyl group of EGCG reduce the ACEi ability, delphinidins-3-*O*-sambubioside and cyanidin-3-*O*-sambubioside chemical structures have primarily ACEi potential.

Studies included *in vivo* cells [50] showed that delphinidin-3-*O*-sambubioside and cyanidin-3-*O*-sambubioside inhibit 43 to 50% the ACE (delphinidin-3-*O*-sambubioside and cyanidin-3-*O*-sambubioside *vs*. control, and 30% less than captopril), furthermore, anthocyanins interfered in the RAS reductive process (RT-qPCR mARN of ACE and renin were analyzed), reducing 37 to 52% the rARN expression for renin. To test the clinic effect of anthocyanins of HS, Nurfaradilla et al. [53] blocked the left renal artery of mice (2KlC hypertension) and treated them with HS extracts (30 mg/200 g BW), captopril, and captopril+HS mixtures; HS extracts reduced the systolic blood pressure 17% (average 150 *vs*. 88, and 80, control *vs*. HS, and captopril), although captopril and HS reduced the renin ad AngII in plasma, HS reduced the ACE activity (1.5 μmol/mL/min control *vs*. 0.40 μmol/mL/min HS, *vs*. 0.30 μmol/mL/min captopril).

Other potential pharmacological properties of HS antioxidants are anti-hypercholesterolemia, antipyretic, antibacterial, antiviral, and anthelminthic [13, 54].

4. The effect of the anthocyanins in ruminant diets and their productive behavior

4.1. Effects of anthocyanins in ruminal digestibility, volatile fatty acids, and potential methane gas emissions. The ability of antioxidants to maintain its activities in ruminal environment, and the molecules abilities to reach the bowel without major modifications. Although some *in vitro* studies show no differences among ruminal gas production and degradability [30, 31, 55], however anthocyanins can improve the ruminal antioxidant potential [30, 31]. Some flavonoids (*e.g*. tannins) have effects on ruminal microbiota [17, 45], modifying the gas production kinetics and the volatile fatty acids (VFA) proportions.

The chemical structure, distribution, and elimination of flavonoids affect the interaction and/or synergism between them and the ruminal microbiota. Although all the effects of anthocyanins in rumen remain unclear, antioxidant and antimicrobial activities of HS are related to the reduction of methane and N-ammonia (CH_4 and NH_3) caused by the changes of the by-products that affect the methanogenic microorganisms growth [45, 47].

Some no desirable antioxidant effects are the reduction of the endogenous fibrolytic enzymes activities, and thereby the potential fiber digestibility and protein absorption [57]. In addition, as other polyphenols sources, some HS components with high-lignin contents have low DM digestibility (DMD), however, antioxidants can modify and improve the biohydrogenation of fatty acids [57] and increase the milk and meat’ polyunsaturated fatty acids (PUFA) [58].

4.2. Effect of anthocyanins after rumen. Some polyphenols are hydrolyzed and transformed through endogenous enzymatic activities and ruminal bacteria [59], therefore the secondary metabolites cross through the ruminal epithelium and the non-absorbed are bio-converted in the small bowel (as it occurs in monogastric) [59] and pass to the bloodstream [34, 57, 60, 61] to deposit in tissues [45, 60].

Anthocyanins can improve the blood plasma resistance to oxidation [32, 62]. Cyanidin-3-*O*-sambubioside and delphinidin-3-*O*-sambubioside can be deposit in lung, cardiac, renal, and hepatic tissues [46], suggesting that anthocyanins can improve the meat and milk antioxidant potential. In addition to the improvement of biohydrogenation of

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 17 May 2021 doi:10.20944/preprints202105.0366.v1

fatty acids in the ruminal environment, anthocyanins increase the animal products to human feed.

Although the milk yield and fat milk have improvements have not been related to anthocyanin addition in ruminant diets [32], the potential clinical effects of delphinidin-3-*O*-sambubioside and cyanidin-3-*O*-sambubioside of HS on RAS, could as in humans, interfere in the homeostatic balance of ruminants affecting the milk yield [63].

Although the reports about the potential effects of HS anthocyanins on fertility parameters are not consistent, other sources of polyphenols, as coffee can improve the semen quality [64] but could reduce the fertilization rates even when progesterone, estradiol, and follicle-stimulating hormone (FSH) remain constant [61]. In contrast, other types of antioxidants as selenium and a-tocopherol might increase some reproductive parameters [27]. Therefore, further studies could be focused on the effect of HS anthocyanins on estrous, and milk and meat production.

5. Effects of HS anthocyanins milk and meat shelf-life

Besides the positive effects of increasing the meat and milk antioxidants on human welfare, anthocyanins could increase the shelf-life of animal products [65-67]. Overall, polyphenols avoid lipids and proteins oxidation (hyper-peroxides, aldehydes, and ketones), autolysis, and microbial pollution [28, 68-70].

6. *Hibiscus sabdariffa* L. by-products

As with other agricultural wastes and by-products the inclusion, of the seeds, stalks, and leaves of HS might reduce the economic and environmental livestock costs [71-73], in addition, optimal inclusion of by-products and wastes in balanced ruminant diets should not have negative effects on animal productive behavior [67, 74-76].

The phenolic and antioxidant activities of HS seeds have been previously assayed and resulted similar or better than those in calyxes [77], but the comparison of the potential effects of seeds with calyxes should be assayed in ruminal liquid, including a test to interpret the ruminal microorganisms-fibrolytic enzymes with the feedstuff' cell walls.

In average, HS seeds have: crude protein (CP), 27.9±10 g/100 g of dry matter (DM); fat, 18.8±8.6 g/100 g DM; crude fiber (CF), 16.8±11.1 g/100 DM; and ashes, 5.86±3.2 g/100 DM (74, 78) (Table 1).

Table 1. *Hibiscus sabdariffa* L. chemical composition.

Authors	DM	CP	EE	CF	Ashes
	g/100 g	g/100 DM			
Maffo et al. [74]	90.0	22.0	22.0	20.0	6.1
Wang et al. [88]	NR	NR	18.0	NR	NR
Ismail et al. [89]	90.0	33.5	22.1	18.3	NR
Shaheen y El-Nakhlawy [90]*	NR	31.4	23.2	4.29	5.5
Udayasekhara [91]**	92.4	20.6	21.0	41.1	5.4
Beshir y Babikier [84]	96.6	30.3	11.1	5.1	5.6
Fagbrnhro [2]	92.6	39.4	6.1	17.7	11.4
Jínez et al. [92]	92.5	20.6	18.0	23.7	6.7
Kwari et al. [93]	NR	38.6	NR	13.5	NR
Mukhtar [94]	91.8	21.4	17.4	12.0	5.3
Soriano y Tejeda [95]	92.7	24.8	17.8	22.9	1.6

doi:10.20944/preprints202105.0366.v1

Anhwange et al. [96]	94.0	19.8	28.0	6.3	5.6
Tounkara et al. [81]	91.8	27.3	20.8	NR	4.5

DM, dry matter; CP, crude protein; EE, ether; CF, crude fiber; NR, no reported; *Average of three varieties; **Average from two varieties.

CP content of HS seeds is comparable to the soybean and canola seeds (79), their fatty acids are primarily oleic and linoleic (n-9 and n-6) (37.68±1.10% and 34.14±1.25%) (Table 2) (10, 80, 81), and its DM and CP *in situ* degradability had been similar to sunflower and peanut seeds [82].

Table 2. Proportion of fatty acids in *Hibiscus sabdariffa* L. seeds and calyxes.

	Seeds		Calyxes
	Tounkara et al. [81]	Mahmoud et al. [80]	Jabeur et al. [11]
Saturated fatty acids (%)			
Miristic (C14:0)	0.21	0.26	1.24 ± 0.01
Palmitic (C16:0)	19.21	20.52	27.73 ± 0.02
Estearic (C18:0)	5.13	5.79	4.46 ± 0.01
Araquidonic (C20:0)	0.67		1.02 ± 0.05
Polyunsaturated fatty acids (%)			
Palmitoleic (C16:1)	0.36		1.32 ± 0.04
Oleic (C18:1)	36.9	38.46	9.1 ± 0.1
Linoleic (C18:2)	35.02	33.25	32.65 ± 0.07
α-linoleic (C18:3)	1.85	1.69	15.76 ± 0.04

Substituting 75% of total CP might not negatively affect animal performance [78]. Previously, the inclusion of ≤25% of the total DM of sheep' diets with HS seeds increased the final body weight and carcass proportion [83], and in other studies, adding 10-20% of HS seeds improved the organoleptic and quality fatty acids properties of sheep meat [84].

Overall, increase the long-chain fatty acids (n-3, n-6, and n-9) in ruminant diets improve the fatty acids composition of milk and meat [85-87].

7. Limitations and perspectives

The relationship among potential antioxidant activities of calyxes, seeds, and stalks anthocyanins of HS with the ruminal microbiota and fibrolytic enzymes remain unknown. In comparison to the studies included in the present review that tested other polyphenols in the ruminal environment, hypothetically positive effects of HS anthocyanins would be the potential reduction of CH_4, and on the fatty acids biohydrogenation process, but also it could reduce the potential fiber degradability [45, 47, 57, 58].

Since antioxidants have a potential reduction of AOPP related with the milk yield improvement, the available information about biochemical and RAS changes promoted by delphinidin-3-*O*-sambubioside and cyanidin-3-*O*-sambubioside of HS [52, 63] could be considered in further *in vivo* studies to find inclusion doses that would improve the composition of the antioxidant and fatty acids and milk yield. However, optimal inclusion should avoid potential negative effects on animal performance and reproductive parameters.

 doi:10.20944/preprints202105.0366.v1

8. Conclusions

The excess of ROS unbalances the oxide-reductive potential in ruminants fed with high-grain diets, exposed to bacterial, viral, and helminthic diseases, and to excessive metabolic exigency and heat stress. HS contain flavonoids primarily classified as anthocyanins which are mainly cyanidin-3-*O*-sambubioside and delphinidin-3-*O*-sambubioside show specific actions on RAS regulation, increasing the ACEi action and reducing the expression of renin genes. In ruminal environment, they can reduce methanogens microorganisms, and interact with substrates, fibrolytic microbiota and enzymes affecting the fiber degradability and the lipids biohydrogenation, which might change the animal performance and the quality of milk and meat lipids. After rumen, anthocyanins are absorbed in small bowel and cross to bloodstream improving the blood resistance to oxidation, and they can be deposited in tissues to increase the milk and meat yields and antioxidant potential. Further studies about the specific action of cyanidin-3-*O*-sambubioside and delphinidin-3-*O*-sambubioside on RAS in ruminants would be useful to understand their potential effects on milk yield, besides this, HS antioxidants should be analyzed on the ruminant reproductive parameters. Although the HS seeds antioxidant effects remain unknown, including HS seeds had not negative affected the ruminant productive behavior but had improved the body weight gain and the fatty quality and proportion in meat.

Author Contributions: Present study was conceptualized, designed, and directed by D.N. Tirado-González. Although all authors were actively involved in data collection, analysis, and discussion process, R. Lazalde-Cruz and M.I. Carrillo-Díaz were responsible to inclusion criteria; L.A. Miranda-Romero and G. Tirado-Estrada were responsible of the formal analysis of results; G.D. Mendoza-Martínez, and A. Lara-Bueno analyzed the validity of derived conclusions; D.N. Tirado González, visualization, and wrote the draft paper. All authors reviewed and edited the final version and have read and agreed with the final decision of submitting and publishing the manuscript.

Acknowledgments: Thank to Universidad Autónoma Chapingo for the technical support.

Conflicts of Interest: Authors declare no conflict of interest. Financial funding sources had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript, or in the decision to publish the results.

References

1. Sáyago, A. S. G.; Arranz, S.; Serrano, J.; Goñi, I. Dietary fiber content and associated antioxidant compounds in Roselle flower (*Hibiscus sabdariffa* L.) beverage. *J Agric Food Chem* **2007**, *55(19), 7886-7890.*
2. Fagbenhro, O. A. Soybean meal substitution with Roselle (*Hibiscus sabdariffa* L.) seed meal in dry practical diets for fingerlings of the African Catfish, *Clarias gariepinus* (Burchell 1822). *J Anim Vet Adv* **2005**, *4(4), 473-477.*
3. SAGARPA. Secretaria de Agricultura, Ganadería, Desarrollo Rural, Pesca y Alimentación. Plan rector nacional sistema producto jamaica. Comité nacional sistema producto jamaica. **2012**. Available online: *https://docplayer.es/8913342-Plan-rector-del-sistema-producto-jamaica.html.*
4. Ariza, F. R.; Serrano, A. V.; Navarro, G. S.; Ovando, C. M.; Vázquez, G. E.; Barrios, A. A.; Michel-Aceves, A. C.; Guzman-Maldonado, S. H.; Otero-Sanchez, M. A. Variedades mexicanas de jamaica (*Hibiscus sabdariffa* L.) 'alma blanca' y 'rosalíz' de color claro, y 'cotzaltzin' y 'tecoanapa' de color rojo. *Rev Fitotec Mex* **2014**, *37(2),181–185.*
5. Aragón, G. A.; Torija, T.; Avelleira, C. R.; Tapia, R.; Contreras, M. I. R.; López, O. J. F. Control de plagas de la jamaica (*Hibiscus sabdariffa L.*) con *Gliricidia sepium* (Jacq.) en Chiautla de Tapia, Puebla. *Rev AIA* **2008**, *12(3), 33-42.*
6. SIAP. Servicio de Información Agroalimentaria y Pesquera. Producción agrícola **2018**. Available at *http://nube.siap.gob.mx/cierre_agricola/.*
7. FAO. Food and Agriculture Organization of the United Nations. *Hibiscus:* Post-harvest Operations. INPhO- Post-harvet Compendium. Roma, Italia. **2004**. Available online: *http://www.fao.org/fileadmin/user_upload/inpho/docs/Post_Harvest_Compendium_-_Hibiscus.pdf*
8. Cid, O. S.; Guerrero, B. J. A. Propiedades funcionales de la Jamaica (*Hibiscus sabdariffa L.*). *Rev TSIA* **2012**, *6 (2), 47-63.*
9. Da-Costa, R. I.; Bonnlaender, B.; Sievers, H.; Pischel, I.; Heinrich, M. *Hibiscus sabdariffa L.* A phytochemical and pharmacological review. *Food Chem* **2014**, *165, 424–443.*
10. Rustan, A. C.; Devron, C. Fatty acids: structures and properties. *Encycl Life Sci* **2005**, *1-7*.
11. Jabeur, I.; Pereira, E.; Barros, L.; Calhelha, R. C.; Sokovic, M.; Oliveira, M. B.; Ferreira, I. C. F. R. *Hibiscus sabdariffa* L. as a source of nutrients, bioactive compounds and colouring agents. *Food Res Int* **2017**, *100, 717–723.*

12. Formagio, A. S. N.; Ramos, D. D.; Vieira, M. C.; Ramalho, S. R.; Silva, M. M.; Zárate, N. A. H.; Foglio, M. A.; Calvalho, J. E. Phenolic compounds of *Hibiscus sabdariffa* and influence of organic residues on its antioxidant and antitumoral properties. *Braz. J. Biol* **2015**, *75(1), 69-76.*
13. Ali, B. H.; Wabel, N. A.; Bluden, G. Phytochemical, pharmacological and toxicological aspects of *Hibiscus sabdariffa L.*: A review. *Phytother Res* **2005**, *19(5), 369-375.*
14. Bergmeier, D.; Berres, P. H. D.; Filippi, D.; Bilibio, D.; Bettiol, V. R.; Priamo, W. L. Extraction of total polyphenols from hibiscus (*Hibiscus sabdariffa* L.) and waxweed /"sete-sangrias " (*Cuphea carthagenensis*) and evaluation of their antioxidant potential. *Acta Sci Technol* **2014**, *(36), 545-551.*
15. Borrás-Linares, I.; Fernández-Arroyo, S.; Arráez-Roman, D.; Palmeros-Suárez, P. A.; Del Val-Díaz, R.; Andrade-Gonzáles, I.; Fernández-Gutiérrez, A.; Gómez-Leyva, J. F.; Segura-Carretero, A. Characterization of phenolic compounds, anthocyanidin, antioxidant and antimicrobial activity of 25 varieties of Mexican Rosselle (*Hibiscus sabdariffa*). *Ind Crop Prod* **2015**, (69), 385-394.
16. Pour, P. M.; Fakhri, S.; Asgary, S.; Farzaei, M. H.; Echeverría, J. The signaling pathways, and therapeutical targets of antiviral agents: Focusing on the antiviral approaches and clinical perspectives of anthocyanin in the management of viral diseases. *Front Pharmacol* **2019**, *10, No. 1207.*
17. Vasta, V.; Daghio, M.; Cappucci, A.; Buccioni, A.; Serra, A.; Viti, C.; Mele, M. Invited review: Plant polyphenols and rumen microbiota responsable for fatty acid biohydrogenation, fiber digestion, and methane emission: Experimental evidence and methodological approaches. *J Dairy Sci* **2019**, *(102), 3781-3804.*
18. Guardiola, S.; Mach, N. Potencial terapéutico del *Hibiscus sabdariffa*: una revisión de las evidencias científicas. *Endocrinol Nutr* **2014**, *61(5), 274-295.*
19. Hygreeva, D.; Pandey, M. C.; Radhakrishna, K. Potential applications of plant-based derivatives as fat replacers, antioxidants and antimicrobials in fresh and processed meat products. *Meat Sci* **2014**, *98,47–57.*
20. Agüero, M.; Segura, C.; Parra, J. Análisis comparativo de compuestos fenólicos totales y actividad antioxidante de cuatro marcas de tisanas de *Hibiscus sabdariffa* (Malvaceae) comercializadas en Costa Rica. *Uniciencia* **2014**, *28(1), 34-42.*
21. Satúe-Gracia, M. T.; Heinonen, M.; Frankel, E. N. Anthocyanins as Antioxidants on Human Low-Density Lipoprotein and Lecithin−Liposome Systems. *J. Agric. Food Chem* **1997**, *45(9),3362–3367.*
22. Kähkönen, M. P.; Hopia, A. I.; Vuorela, H. J.; Rauha, J. P.; Pihlaja, K.; Kujala, T. S.; Heinonen, M. Antioxidant Activity of Plant Extracts Containing Phenolic Compounds. *J Agric Food Chem* **1999**, *47(10), 3954-3962.*
23. Galicia-Flores, L. A.; Salinas-Moreno, Y.; Espinoza-García, B. M.; Sánchez-Feria, C. Caracterización fisicoquímica y actividad antioxidante de extractos de jamaica (*Hibiscus Sabdariffa L.*) Nacional e importada. *Rev Chapingo Ser Hortic* **2008**, *14(2), 121-129.*
24. Borrás, L. A.; Salinas-Moreno, Y.; Espinoza-García, B. M.; Sánchez-Feria, C. Caracterización fisicoquímica y actividad antioxidante de extractos de jamaica (*Hibiscus Sabdariffa L.*) Nacional e importada. *Rev Chapingo Ser Hortic* **2008**, *14(2), 121-129.*
25. Sindi, H. A.; Marshall, L. J.; Morgan, M. R. A.; Comparative chemical andbiochemical analysis of extracts of *Hibiscus sabdariffa*. *Food Chem* **2014**, *164, 23–29.*
26. Chu, C.; Deng, J.; Man, Y.; Qu, Y. Green Tea Extracts Epigallocatechin-3-gallate for Different Treatments. *Biomed Res Int* **2017**, id *5615647*.
27. Chauhan, S. S.; Celi, P.; Ponnampalam, E. N.; Leury, B. J.; Lui, F.; Dunshea, F. R. Antioxidant dynamics in the live animal and implications for ruminant health and product (milk/meat) quality: role of vitamin E and selenium. *Anim Prod Sci* **2014**, *54,1525-1536.*
28. Cuhna, L. C. M.; Monteiro, M. L.; Lorenzo, J. M.; Munekata, P. E.; Muchenje, V.; Carvalho, F. A.; Conte-Junior, C. A. Natural antioxidants in processing and storage stability of sheep and goat meat products. *Food Res Int* **2018**, *111(2018), 379–390.*
29. Lorenzo, J. M.; Pateiro, M.; Domínguez, R.; Barba, F. J.; Putnik, P.; Bursac, K. D.; Shpigelman, A.; Granato, D.; Franco, D. Berries extracts as natural antioxidants in meat products: A review. *Food Res Int* **2018**, *106, 1095–1104.*
30. Hosoda, K.; Sasahara, H.; Matsushita, K.; Tamura, Y.; Miyaji, M.; Matsuyama, H. Anthocyanin and proanthocyanidin contents, antioxidant activity, and *in situ* degradability of black and red rice grains. *Asian-Aust J Anim Sci* **2018**, *31(8), 1213-1220.*
31. Tian, X.; Paengkoum, P.; Paengkoum, S.; Thongpea, S.; Ban, C. Comparison of forage yield, silage fermentative quality, anthocyanin stability, antioxidant activity, and *in vitro* rumen fermentation of anthocyanin-rich purple corn (*Zea mays* L.) stover and sticky corn stover. *J Integra Agri* **2018,** *17(9), 2082-2095.*
32. Hosoda, K.; Eruden, B.; Matsuyama, H.; Shioya, S. Effect of anthocyanin-rich corn silage on digestibility, milk production and plasma enzyme activities in lactating dairy cows. *Anim Sci J* **2012a**, *83(6), 453-459.*
33. Theodorou, M. K.; Kingston-Smith, A. H.; Winters, A. L.; Lee, M. R. F.; Minchin, F. R.; Morris, P.; MacRae, J. Polyphenols and their influence on gut function and health in ruminants: a review. *Environ Chem Lett* **2006**, *(4), 121-126.*
34. Gladine, C.; Rock, E.; Morand, C.; Bauchart, D.; Durand, D. Bioavailability and antioxidant capacity of plant extracts rich in polyphenols, given as a single acute dose, in sheep made highly susceptible to lipoperoxidation. *Br J Nutr* **2007**, *(98), 691-701.*

 doi:10.20944/preprints202105.0366.v1

35. Olagaray, K. E.; Bradford, B. J. Plant flavonoids to improve productivity of ruminants –A review. *Anim Feed Sci Tech* **2019**, *(251), 21-36*.
36. Ametaj, B. N.; Zebeli, Q.; Saleem, F.; Psychogios, N.; Lewis, M. J.; Dunn, S. M.; Xia, J.; Wishart, D. S. Metabolomics reveals unhealthy alterations in rumen metabolism with increased proportion of cereal grain in the diet of dairy cows. *Metabolomics* **2010**, *(6), 583-594.*
37. Cell, P.; Gabal, G. Oxidant/antioxidant balance in animal nutrition and health: the role of protein oxidation. *Front Vet Sci* **2015**, *2 (48), 1-13.*
38. Estévez, M.; Li, Z.; Soladoye, O. P.; Van-Hecke, T. Health risks of food oxidation. *Adv Food Nutr Res* **2017**, *(82), 45-81*.
39. Daley, C. A.; Abbot, A.; Doyle, P.; Nader, G. A.; Larson, S. A review of fatty acid profiles and antioxidant content in grass-fed and grain-fed beef. *Nut J* **2010**, *9(10), 1-12.*
40. He, Y.; Tan, D.; Mi, Y.; Zhou, Q.; Ji, S. Epigallocatechin-3-gallet attenuates cerebral cortex damage and promotes brain regeneration in acrylamide-treated rats. *Food Funct* **2017**, *8(6), 2275-2282.*
41. Ortíz-Romero, P.; Borralleras, C.; Campuzano, V. Epigallocatechin-3-gallate improves cardiac hypertrophy and short-term memory deficits in a Williams-Beuren syndrome mouse model. *PLoS One* **2018**, *13(3), e0194476.*
42. Wang, Y.; Wang, B.; Du, F.; Su, X.; Sun, G.; Zhou, G.; Bian, X.; Liu, Na. Epigallocatechin-3-gallate attenuates ureteral obstruction-induced renal interstitial fibrosis in mice. *J Histochem Cytochem* **2015**, *63(4), 270-279.*
43. Palabiyik, S. S.; Dincer, B.; Cadirici, E.; Cinar, E.; Gundogdu, C.; Polat, B.; Yayla, M.; Halici, Z. A new update for radio-contrast-induced nephropathy aggravated with glycerol in rats: the protective potential of epigallocatechin-3-gallate. *Ren Fail* **2017,** *39(1), 314-322.*
44. Chen, L. L.; Xu, Y. Epigallocateichin gallate attenuates uric acid-induced injury rat renal interstitial fibroblasts NRK-49F by up-regulation of miR-9. *Eur Rev Pharmacol Sci* **2018**, *(21), 7458-7469*.
45. Klantar, M. The importance of flavonoids in ruminant nutrition. *Arch Animal Husb & Dairy Sci* **2018**, *1(1), 1-4.*
46. Sandoval-Ramírez, B. A.; Catalán, U.; Fernández-Castillejo, S.; Rubio, L.; Macia, A.; Sola, R. Anthocyanin tissue bioavailability in animals: Possible implications for human health. A systematic review. *J Agric Food Chem* **2018**, *66, 11531-11543.*
47. Xiao, X.; Du, C.; Yan, Z.; Shi, Y.; Duan, H.; Ren, Y. Inhibition of necroptosis attenuates kidney inflammation and interstitial fibrosis induced by unilateral ureteral obstruction. *Am J Nephrol* **2017**, *46(2), 131-138.*
48. Kakuta, Y.; Okumi, M.; Isaka, Y.; Tsutahara, K.; Abe, T.; Yazawa, K.; Ichumaru, N.; Matsumura, K.; Hyon, S.; Takahara, S.; Nonomura, N. Epigallocatechin-3-gallate protects kidneys from ischemia rerperfusion injury by HO-1 upregulation and inhibition of macrophage infiltration. *Transpl Int* **2011**, *24(5), 514-522*.
49. Kim, Hae-Suk.; Quon, M. J.; Jeong-A, Kim. New insights into the mechanisms of polyphenols beyond antioxidant properties; lessons from the green tea polyphenol, epigallocatechin 3-gallate. *Redox Biol* **2014**, *2, 187-195.*
50. Parichatikanond, W.; Pinthong, D.; Mangmool, S. Blockade of the renin-angiotensin system with delphinidin, cyanin, and quiercetin. *Planta Med* **2012**, *78, 1626-1632.*
51. Nakamura, K.; Shimizu, T.; Yanagita, T.; Nemoto, T.; Taniuchi, K.; Shimizu, S.; Dimitriadis, F.; Yawata, T.; Higashi, Y.; Ueba, T.; Saito, M. Angiotensin II acting on brain AT1 receptors induces adrenaline secretion and pressor responses in the rat. *Sci Rep* **2014**, *4, 7248.*
52. Guerrero, L.; Castillo, J.; Quiñones, M.; Garcia-Vallvé, S.; Arola, L.; Pujadas, G.; Muguerza, B. Inhibition of angiotensin-converting enzyme activity by flavonoids: structure-activity relationship studies. *PLoS One* **2012**, *7(11), e49493.*
53. Nurfaradilla, S. A.; Saputri, F. C.; Harahap, Y. Effects of *Hibiscus Sabdariffa* calyces aqueous extract on the antihypertensive potency of captopril in the two-kidney-one-clip rat hypertension model. *Evid Based Complement Alternat Med* **2019**, *Id. 9694212.*
54. Nileeka, B. W.; Rupasinghe, H. P. Plant flavonoids as angiotensin converting enzyme inhibitors in regulation of hypertension. *Funct Food Health Dis* **2011**, *1(15), 172-188.*
55. Elahi, M. Y.; Rouzbehan, Y.; Rezaee, A. Effects of phenolic compounds in three oak species on *in vitro* gas production using inoculums of two breeds of indigenous Iranian goats. *Anim Feed Sci Tech* **2012**, *176, 26-31.*
56. Klantar, M. The importance of flavonoids in ruminant nutrition. *Arch Animal Husb & Dairy Sci* **2018**, *1(1): id.000504.*
57. Corredeu, F.; Lunesu, M. L.; Buffa, G.; Atzori, A. S.; Nudda, A.; Battacone, G.; Pulina, G. Can agro-industrial by-products rich in polyphenols be advantageously used in feeding and nutrition of dairy small ruminants? *Animal* **2020**, *10(1), 131.*
58. Morales, R.; Ungerfeld, E. M. Use of tannins to improve fatty acids profile of meat and milk quality in ruminants: A review. *Chilean J Agric Res* **2015**, *75(2), 239-248.*
59. Gessner, D. K.; Ringseis, R.; Eder, K. Potential of plant polyphenols to combat oxidative stress and inflammatory processes in farm animals. *J Anim Physiol Anim Nutr* **2016**, *101(4), 605-628.*
60. Castillo, C.; Hernández, J.; López-Alonso, M.; Miranda, M.; Benedito, J. L. Values of plasma lipid hydroperoxides and total antioxidant status in healthy dairy cows: preliminary observations. *Archiv fur Tierzucht* **2003**, *46(1), 227-233.*
61. Salinas-Rios, T.; Sánchez-Torres-Esqueda, M. T.; Díaz-Cruz, A.; Cordero-Mora, J. L.; Cárdenas-León, M.; Hernández-Bautista, J.; Nava-Cuellar, C.; Nieto-Aquino, R. Oxidative status and fertility of ewes supplemented coffee pulp during estrous synchronization and early pregnancy. *Rev Col Cienc Pecu* **2016,** *29, 255-263.*

62. Hosoda, K.; Miyaji, M.; Matsuyama, H.; Haga, S.; Ishizaki, H.; Nonaka, K. Effect of supplementation of purple pigment from anthocyanin-rich corn (*Zea mays L.*) on blood antioxidant activity and oxidation resistance in sheep. *Livest Sci* **2012b**, *145, 266-270.*
63. La Rosa, M.; Kechichian, T.; Olson, G.; Saade, G.; Prewit, E. B. Lactation leads to modifications in maternal Renin-Angiotensin System in later life. *Obstet Gynecol* **2020**, *27, 260-266.*
64. Janegrad, P. A.; Kia, H. A. Moghaddam G, Ebrahimi M. Evaluating caffeine antioxidant properties on Ghezel ram sperm quality after freeze-thawing. *Revue Méd Vét* **2018**, *169(10-12), 233-240.*
65. Zhang, M.; Hettiarachchy, N. S.; Horax, R.; Kannan, A.; Praisoody, A. M. D.; Muhundan, A.; Mallangi, C. R. Phytochemicals, antioxidant and antimicrobial activity of *Hibiscus sabdariffa*, *Centella asiatica*, *Moringa oleifera* and *Murraya koenigii* leaves. *J Med Plant Res* **2011**, *5(30), 6672-6680.*
66. Valenzuela, V. C.; Pérez, P. M. Actualización en el uso de antioxidantes naturales derivados de frutas y verduras para prolongar la vida útil de la carne y productos cárneos. *Rev Chil Nutr* **2016**, *43(2),188-195.*
67. Nikmaram, N.; Budaraju, S.; Barba, F. J.; Lorenzo, J. M.; Cox, R. B.; Mallikarjunan, K.; Roohinejad, S. Application of plant extracts to improve the shelf-life, nutritional and health-related properties of ready-to-eat meat products. *Meat Sci* **2018**, *145, 245-255.*
68. Karre, L.; Lopez, K.; Getty, K. J. Natural antioxidants in meat and poultry products. *Meat Sci* **2013,** *94, 220–227.*
69. Falowo, A. B.; Fayemi, P. O.; Muchenje, V. Natural antioxidants against lipid–protein oxidative deterioration in meat and meat products: A review. *Food Res Int* **2014**, *64, 171–181.*
70. Jiang, J.; Xiong, Y. L. Natural antioxidants as food and feed additives to promote health benefits and quality of meat products: A review. *Meat Sci* **2016**, *120, 107–117.*
71. McGinn, S. M.; Beauchemin, K. A.; Coates, T.; Colombatto, D. Methane emissions from beef cattle: effects of monensin, sunflower oil, enzymes, yeast, and fumaric acid. *J Anim Sci* **2004**, *82, 3346–3356.*
72. Beauchemin, K.; Kreuzer, M.; O´Mara, F. O.; McAllister, T. A. Nutritional management for enteric methane abatement: *A review. Aust J Exp Agric* **2008**, *48, 21-27.*
73. Knapp, J. R.; Laur, G. L.; Vadas, P. A.; Weiss, W. P.; Tricarico, J. M. Invited review: enteric methane in dairy cattle production: quantifying the opportunities and impact of reducing emissions. *J Dairy Sci* **2014**, *97, 3231–3261.*
74. Maffo, T.; Agbor, E. E.; Mekoudjou, N. H.; Kengne, S. C. N.; Gouado, I. Proximate and mineral composition, protein quality of *Hibiscus sabdariffa* L. (roselle) seeds cultivated in two agroecological areas in Cameroon. *Int J Nutr Food Sci* **2014**, *3(4), 251-258.*
75. Mora, A. M. E.; Tirado-González, D. N.; Guevara-Lara, F.; Jáuregui-Rincón, J.; Larios-González, R.; Tirado-Estrada, G. Calidad nutricional del bagazo de manzana ensilado con fuentes nitrogenadas orgánicas e inorgánicas. *Rev Mex Cienc Agríc* **2018**, *9(1), 229-235.*
76. Tirado-Estrada, G.; Ramos-Mijangos, L. M.; Miranda-Romero, L. A.; Tirado-González, D. N.; Salem, A. Z. M.; Mlambo, V.; Medina-Cuellar, S. E.; González-Reyes, M.; Barababosa-Pliego, A. Potential impacts of dietary *Lemna gibba* supplements in a simulated ruminal fermentation system and environmental biogas production. *J Clean Prod* **2018**, *18, 555-561.*
77. Mohd-Esa, N.; Shin, H. F.; Ismail, A.; Lye, Y. C. Antioxidant activity in different parts of roselle (*Hibiscus sabdariffa* L.) extracts a potential exploration of the seeds. *Food Chem* **2010**, *122, 1055-1060.*
78. Sulliman, G. M.; Babiker, S. A.; Eichinger, H. M. Effect of Hibiscus seed-based diet on chemical composition, carcass characteristics and meat quality traits of cattle. *Indian J Anim Res* **2017**, *51(4), 694-699.*
79. Bellaloui, N.; Bruns, H. A.; Abbas, H. K.; Mengisty, A.; Fisher, D. K.; Reddy, K. N. Agricutural practices altered soybean protein, oil, fatty acids, sugars, and minerals in the Midsouth USA. *Front Plant Sci* **2015**, *6(31), 1-14.*
80. Mahmoud, A. A.; Selim, K. A.; Abdel-Baki, M. R. Physico-chemical and oxidative stability characteristics of roselle (*Hibiscus sabdariffa* L.) seed oil as by-product. *Egypt J Appl Sci* **2008**, *23(7), 247-257.*
81. Tounkara, F.; Amadou, I.; Le, G.; Shi, Y. Effect of boiling on the physicochemical properties of Roselle seeds (*Hibiscus sabdariffa* L.) cultivated in Mali. *African J Biotech* **2011**, *10(79),18160-18166.*
82. Rahman, A. A.; Salih, A. M.; Turki, I. Y.; Abdulatee, S. O.; Elbasheir, O. M.; Magboul, M. M. Determination of dry matter and crude protein degradability of Roselle seed (*Hibiscus sabdariffa*) using bags nylon technique. *Sudan J Sci Tech* **2016**, *17(1), 58-65.*
83. Elamin, K. M.; Hassan, H. E.; Abdalla, H. O.; Arabi, O. H.; Tameem, E. A. A. Effect of feeding crushed roselle seed (*Hibiscus sabdariffa* L.) on carcass characteristics of sudan desert sheep. *Asian J Anim Sci* **2012**, *6(5), 240-248.*
84. Beshir, A. A.; Babiker, S. A. The effect of feeding diet with graded levels of roselle (*Hibiscus sabdariffa*) seed on carcass characteristics and meat quality of sudan desert lamb. *Res J Animal Vet Sci* **2009**, *4, 35-44.*
85. Christensen, R. A.; Drackley, J. K.; LaCount, D. W.; Clark, J. H. Infusion of four long-chain fatty acids mixtures into de abomasum of lactating dairy cows. *J Dairy Sci* **1994**, *77(4), 1052-1069.*
86. Martínez-Borraz, A.; Moya-Camarena, S. Y.; González-Ríos, H.; Hernández, J.; Pinelli-Saavedra, A. Conjugated linoleic acid (CLA) content in milk from confined Holstein cows during summer months in northwestern Mexico. *Rev Mex Cienc Pecu* **2010,** *1(3), 221-235.*
87. Hernández, A.; Kholif, A. E.; Lugo-Coyote, R.; Elghandour, M. M. Y.; Cipriano, M.; Rodríguez, G. B.; Odongo, N. E.; Salem, A. Z. M. The effect of garlic oil, xylanase enzyme and yeast on biomethane and carbon dioxide production from 60-d old Holstein dairy calves fed a high concentrate diet. *J Clean Prod* **2017**, 148, 616-623.

88. Wang, M. L.; Morris, B.; Tonnis, B.; Davis, J.; Pederson, G. A. Assessment of oil Content and fatty acid composition variability in two economically important *Hibiscus* species. *J Agric Food Chem* **2012**, *60, 6620–6626*.
89. Ismail A, Khairul IE, Mohd NH. Roselle (*Hibiscus sabdariffa* L.) sedes-nutritional composition, protein quality and health benefits. Global Sci Book Food 2008; 2(1):1-16.
90. Shaheen, M. A.; El-Nakhlawy, F. S.; Al-Shareef, A. R. Roselle (*Hibiscus sabdariffa* L.) seeds as unconventional nutritional source. *African J Biotech* **2012**, *11(41), 821-9824*.
91. Udayasekhara, R. P. Nutrient composition and biological evaluation of mesta (*Hibbiscus sabdariffa*) seeds. *Plant Foods Hum Nutr* **1996**, *49, 27-34*.
92. Jínez, M. T.; Cortés, C. A.; Ávila, G. E.; Casaubon, M. A.; Salcedo, E. R. Efecto de niveles elevados de semilla de jamaica (*Hibiscus sabdariffa*) en dietas para pollos sobre el comportamiento productivo y funcionamiento hepático. *Veterinaria Mexicana* **1998**, *29(1), 35-40*.
93. Kwari, I. D.; Igwebuike, J. U.; Mohammed, I. D.; Diarra, S. S. Growth, haematology and serum chemistry of broiler chickens fed raw or differently processed sorrel (*Hibiscus sabdariffa*) seed meal in a semi-arid environment. *IJSN* **2011**, *2(1), 22-27*.
94. Mukhtar, A. M. The Effect of Feeding Rosella (*Hibiscus Sabdariffa*) Seed on Broiler Chick's Performance. *Res J Anim Vet Sci* **2007**, *2, 21-23*.
95. Soriano, T. J.; Tejada, H. I. Estudio preliminar del valor nutritivo de la semilla de jamaica (*Hibiscus sabdariffa*) para el pollo de engorda. *Técnica Pecuaria Mexicana* **1995**, *33, 48-52*.
96. Anhwange, B. A.; Ajibola, V. O.; Okibe, F. G. Nutritive value and anti-nutritional factors in *Hibiscus sabdariffa*. *J Fish Intl* **2006**, *1(2-4), 73-76*.