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PLASMA N-3 AND N-6 FATTY ACID PROFILES AND THEIR CORRELATIONS TO HAIR COAT SCORES IN HORSES KEPT UNDER MALAYSIAN CONDITIONS

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SUMMARY

A survey was carried out to determine the relationship between plasma fatty acid profiles and hair coat scores of horses kept under Malaysian conditions. Thirty-seven Thoroughbred horses with an average age of seven years were included in this study. The surveyed population comprised 27 geldings and 10 mares from the Kuala Lumpur City Hall stables at Titiwangsa (TTW, n = 7) and Bandar Tun Razak (BTR, n = 7), the Equine Unit, Universiti Putra Malaysia (UPM, n = 7), the Royal Malaysian Police stables (RMP, n = 8) and the Selangor Turf Club (STC, n = 8). Plasma and feed fatty acid profiles were determined using gas chromatography and the hair coat score determined using a seven-point scoring system (1 = worst hair coat condition; 7 = best hair coat condition). Results showed that feed and plasma fatty acids profiles were variable across sampling locations. The n-6 : n-3 ratios in feeds ranged from 6.8 (BTR) to 17.2 (UPM). Only plasma oleic and linolenic acids were different ($P<0.05$) across sampling locations. The STC horses had the best hair coat score (median score = 7, $P< 0.05$). It was found that total n-3 fatty acids were highly correlated with hair coat scores ($\rho = 0.686$, $P<0.01$). A significant inverse correlation between n-6: n-3 ratio and hair coat scores ($\rho = -0.755$, $P<0.01$) was also noted. This meant that increasing plasma n-3 fatty acids and decreasing n-6: n-3 ratios were associated with better hair coat scores in these horses.

Keywords: horses, hair coat, n-3 fatty acids, n-6 fatty acids

INTRODUCTION

Fatty acids are critical for structural, storage and metabolic functions of the mammalian body (Gurr *et al.*, 2002). They are important as an energy source in hormone synthesis or incorporated into cell membranes (Lands, 1992). Fatty acids also serve other functional roles such as those related to structural integrity, regulation of the body functions and modulation of the immune system (O'Keefe, 1998). The essential fatty acids (EFA) are fatty acids that the animal body is not able to synthesise, or at least not adequately, to meet the body's requirement. The EFA are grouped into two families, the n-6 and the n-3 fatty acids. The n-3 and n-6 fatty acids are important in the development and maintenance of healthy skin and hair coat, as well as a concentrated source of energy for high-level performance horses (Cunha, 1991). These fatty acids are important in the maintenance of epidermal barrier function (Thompson, 1992), as phospholipid components of cell membranes and as precursors of a variety of potent, short-lived molecules including the prostaglandins, thromboxanes, leukotrienes and hydroxyeicosatetraenoic acids (Ruzicka and Printz, 1984). The n-3 fatty acids have anti-inflammatory, antithrombotic, antiarrhythmic, hypolipidemic, and vasodilatory properties (Simopoulus, 1999), whereas the n-6 fatty acids, in particular the arachidonic acid, plays a central role in the production of many important pro-inflammatory substances. When tissue is damaged, phospolipase A2 acts to break down membrane phospholipid, providing free arachidonic acid, which can be acted upon by the lipoxygenase and cyclo-oxygenase enzymes to produce substances with potent, predominantly pro-inflammatory properties (Vorhees, 1983). Release of free arachidonic acid is inhibited by glucocorticoids and also by the action of prostaglandins. The lipoxygenase within the pro-inflammatory arachidonic cascade is inhibited by 15-hydroxy-dihomogammalinolenic acid, a metabolite of dihomogammalinoleic acid (Miller *et al.*, 1989). These were the basis of drug treatment in alleviating inflammation of the skin.

The precursor of the n-3 fatty acids, the linolenic acid, is found in vegetable oils such as linseed and cold water marine fishes. Principal sources of the n-6 fatty acids include sunflower oil and corn oil which provide linoleic acid, and evening primrose oil which provides linoleic acid and significant amounts of gamma-linolenic acid. These fatty acids are metabolized to their longer-chain metabolites mainly in the liver. The skin lacked both delta-6 and delta-5 desaturases, although it does have elongase activity (Chapkin *et al.*, 1986). Therefore, it is

dependent on vascular supplies of linoleic, gamma-linoleic and arachidonic acids. The high cell turnover rate of the epidermal layer thus resulted in the skin being particularly vulnerable to n-3 and n-6 fatty acid deficiencies or their imbalances (Lloyd, 1989). Disruption of the n-3 and n-6 fatty acid status of the skin has been known to result in poor skin conditions (Rees *et al.*, 2001). Deficiencies of n-3 and/or n-6 fatty acids can occur in many species of mammals, usually in association with malnutrition or malabsorption (Sherertz, 1986). Essential fatty acid deficiency in most species causes a dry, lusterless hair coat and scaly skin, and may predispose to skin infection. If the deficiency persists, hair loss, edema, and exudation from localised areas of the skin, resulting in moist inflammation may ensue (Lewis, 1996). An increase in the n-3 and n-6 fatty acid levels would improve the hair coat score, indicating a better hair coat condition (Rees *et al.,* 2001). This was due to the fact that n-6 and n-3 fatty acids contribute to cell membrane fluidity and skin health (Gurr *et al.,* 2002), thus modulating hair coat conditions indirectly (Lewis, 1996).

The n-6 and n-3 fatty acid-based oils had been used successfully in the treatment of Queensland's Itch and other skin conditions in horses (Lloyd and Thomsett, 1989; Bond and Lloyd, 1993; Scott *et al.,* 1997). This is not surprising as dietary polyunsaturated fatty acids (PUFA), especially the n-3 fatty acids, show excellent results in treating dermatological conditions in many other animal species and human patients (Bauer, 1994; Andreassi *et al.,* 1997). Therefore, it was hoped that this study would further elucidate the importance of dietary n-3 and n-6 fatty acids in equine skin health. The objectives of this study were to determine the n-3 and n-6 fatty acid profiles of the horse plasma and commonly used horse feeds, and secondly, to ascertain the probable correlations between the n-3 and n-6 fatty acid profiles with the hair coat conditions in horses.

MATERIALS AND METHODS

Thirty-seven Thoroughbred horses with an average age of seven years were included in this survey. The surveyed population comprised 27 geldings and 10 mares, which were evenly distributed among the five predetermined stables within a 30 km radius from Universiti Putra Malaysia, Serdang, Selangor. Seven horses were obtained from each of the Kuala Lumpur City Hall stables at Titiwangsa (TTW, n = 7) and Bandar Tun Razak (BTR, n = 7). Seven horses were from the Equine Unit at the Universiti Putra Malaysia (UPM, n = 7), while both the Royal Malaysian Police (RMP, n = 8) and Selangor Turf Club (STC, n = 8) stables contributed eight horses each. Horses included in this survey were determined to have similar hair coat management and were groomed within five days before the hair coat scoring was performed. Dietary records showed that all the five stables had different diets and dietary managements. The fatty acid profiles of the diet from these locations are shown in Table 1.

Determination of plasma and feed fatty acid profiles

Ten mL of blood was collected from the jugular vein from all horses into ethylenediamine tetra-acetic acid (EDTA) vacutainer tubes. The blood was then centrifuged at 1000 G for 10 min to obtain the plasma. The plasma collected was then kept in a refrigerator at -20°C until total lipid extractions. Feed samples collected from all five locations were ground to pass a one mm sieve and subjected to total lipid extractions in triplicates.

Total fatty acids were extracted from the plasma and feeds using the chloroform-methanol 2:1 (v/v) solvent system according to the Folch method described by Rajion *et al.,* (2001). Forty mL of chloroform-methanol (2:1, v/v) were added to three mL of plasma or 0.5 g of feed sample in a 50 mL stoppered tube. The mixture was shaken, flushed with nitrogen and sealed. The mixture was left to stand for at least twelve hours and was then filtered through a No.1 Whatman filter paper into a separating flask. Five mL of chloroform-methanol (2:1, v/v) was used to wash the lipid residue on the tube and the filter paper. Ten mL of normal saline was then added into the separating flask to facilitate phase separation. The mixture was then left to stand for at least four hours. After complete separation, the lower phase was collected into a round flask bottle and evaporated by rotary evaporation at 70-75°C. The extracted lipids were transferred to methylation tubes with stoppers and chloroform-methanol (2:1, v/v) was added to a final volume of five mL. The lipid extract was then dried and concentrated under nitrogen gas. The fatty acids were transmethylated to fatty acid methyl esters (FAME) using 14 % methanolic boron trifluoride, and separated on a Supelco SP™-2330 fused silica capillary column (30m, 0.25mm ID, 0.20 µm film thickness, Supelco Inc., Bellefonte, PA, USA) in a 5890 Hewlett-Packard Gas-Liquid Chromatograph (Hewlett-Packard Co., Avondale, PA, USA) equipped with a Flame Ionization Detector (FID). Purified nitrogen gas flowing at 40 mL/min was used as the carrier gas. The injector temperature was programmed at 220 °C and the detector at 220°C. The column temperature was set at a range of 100-190°C with temperature increment programmed at a rate of 7.2°C/min to facilitate optimal separation. Identification of the fatty acid methyl esters was based on the comparison of the sample retention times to those of a known fatty acid methyl ester standard (Sigma Chemical Co., St Louis, MO, USA). An internal standardisation method was used to quantify the various fatty acids in the plasma, where a known concentration of heneicosanoic acid (21:0) (Sigma Chemical Co., St Louis, MO, USA) was added to each sample prior to transmethylation.

Table 1: Mean dietary fatty acid compositions by survey locations (as mg/100g feed & % total fatty acids)

	STC (n=3)		UPM (n=3)		BTR (n=3)		TTW (n=3)		RMP (n=3)	
Fatty acids	mg/100g	(%)	mg/100g	(%)	mg/100g	(%)	mg/100g	(%)	mg/100g	(%)
Lauric acid (12:0)	6.8	*(0.2)*	3.2	*(0.1)*	3.1	*(0.1)*	10.1	*(0.5)*	3.6	*(0.1)*
Myristic acid (14:0)	2.1	*(0.0)*	4.8	*(0.1)*	2.3	*(0.1)*	4.7	*(0.2)*	5.8	*(0.2)*
Palmitic acid (16:0)	470.2	*(11.1)*	1006.4	*(21.2)*	445.2	*(17.1)*	360.8	*(17.8)*	531.4	*(16.2)*
Palmitoleic acid (16:1)	7.3	*(0.2)*	92.0	*(1.9)*	433.0	*(16.6)*	77.7	*(3.8)*	72.5	*(2.2)*
Stearic acid (18:0)	175.7	*(4.2)*	133.6	*(2.8)*	57.5	*(2.2)*	47.7	*(2.4)*	75.4	*(2.3)*
Oleic acid (18:1)	1632.4	*(38.6)*	1598.4	*(33.6)*	558.2	*(21.5)*	472.7	*(23.4)*	1162.1	*(35.4)*
Linoleic acid (18:2 n-6)	1767.3	*(41.8)*	1808.3	*(38.1)*	960.9	*(36.9)*	938.6	*(46.4)*	1313.2	*(40.0)*
Linolenic acid (18:3 n-3)	111.4	*(2.6)*	105.0	*(2.2)*	142.1	*(5.5)*	109.4	*(5.4)*	117.0	*(3.6)*
Behenic acid (22:0)	40.4	*(1.0)*	*ND*	*ND*	*ND*	*ND*				
Lignoceric acid (24:0)	13.5	*(0.3)*	*ND*	*ND*	*ND*	*ND*				
Total saturated fatty acids (SFA)	708.7	*(16.8)*	1148.0	*(24.2)*	508.1	*(19.5)*	423.3	*(20.9)*	616.2	*(18.8)*
Total unsaturated fatty acids (UFA)	3518.4	*(83.2)*	3603.7	*(75.8)*	2094.2	*(80.5)*	1598.4	*(79.1)*	2664.8	*(81.2)*
Total PUFA n-3 (or omega-3)	111.4	*(2.6)*	105.0	*(2.2)*	142.1	*(5.5)*	109.4	*(5.4)*	117.0	*(3.6)*
Total PUFA n-6 (or omega-6)	1767.3	*(41.8)*	1808.3	*(38.1)*	960.9	*(36.9)*	938.6	*(46.4)*	1313.2	*(40.0)*
n-6 to n-3 ratio	15.9		17.2		6.8		8.6		11.2	
UFA to SFA ratio	5.0		3.1		4.1		3.8		4.3	

ND – not detected

Hair coat scoring

Hair coat condition of each horse was photographed and evaluated immediately after blood sampling using a seven-point numerical scoring system as follows:

Score 1: Dull, coarse, broken hair distributed all over the body area.
Score 2: Dull, coarse, broken hair distributed on most (60%) of the body area.
Score 3: Dull, coarse, broken hair distributed on about 40% of the body area.
Score 4: Dull, coarse, patches of broken hair on about 20% of the body area.
Score 5: Slightly shiny with a few patches of coarse and broken hair on less than 20 % of the body area.
Score 6: Reasonably smooth and shining hair coat with minimal or no broken hair.
Score 7: Very smooth, shining and healthy looking hair coat.

Statistical analyses

The plasma fatty acid profiles were analysed using the one way analysis of variance (ANOVA) procedure to examine for probable differences due to sampling locations. Significantly different means were then differentiated using the Duncan Multiple Range Test and Least Significant Difference Test. Hair coat scores were analysed using the Kruskal-Wallis H-test. Significant groups were further elucidated using the Q-statistics test. Spearman's Rank Correlation was performed to investigate the relationship between plasma total n-3 fatty acid and n-6 fatty acid levels, and overall hair coat score. All statistical analyses were performed using the SPSS software at 95 % confidence level.

RESULTS

It is evident from Table 1 that feed samples from the surveyed stables had different fatty acid compositions. Ratios of the total n-6 fatty acids to total n-3 fatty acids ranged from 6.8 (BTR) to 17.2 (UPM). This was due to the high concentrations of total n-6 fatty acids coupled with the lowest linolenic acid concentrations in feed samples from UPM. In general, feed samples from STC, UPM and RMP had almost twice the absolute amount of linoleic acid, compared to TTW and BTR as a result of higher levels of grain-based horse pellet inclusion in their diets. The feed from STC also had the highest proportion of total unsaturated fatty acids and consequently the best UFA to SFA ratio at 5.0. The STC diet also had behenic and lignoceric acids, which were not found in the other four feed samples.

Plasma fatty acids

The plasma linolenic acid level of the STC horses was statistically different ($P<0.05$) from TTW, RMP and UPM horses (Table 2). The STC horses had the highest concentration of plasma linolenic acid (6.2 ± 0.3 mg/100 mL), while horses from TTW had the lowest at 3.3 ± 0.7 mg/100 mL. These differences contributed to the significant variation ($P<0.05$) of total plasma n-3 fatty acid concentrations as a result of sampling location in this study (Table 2). Consequently, the total n-6 fatty acid

Table 2: The mean plasma fatty acid compositions by survey locations (as mean + SEM mg/100mL plasma & % total fatty acids)

	STC (N=8)		UPM (N=7)		BTR (N=7)		TTW (N=7)		RMP (N=8)	
Fatty acids	(mg/100g)	%	(mg/100g)	%	(mg/100g)	%	(mg/100g)	%	(mg/100g)	%
Palmitic acid^{ns} (16:0)	26.2 ± 1.2	13.5	27.5 ± 1.6	*14.5*	30.7 ± 1.4	15.5	25.0 ± 2.0	*13.3*	28.2 ± 1.1	*14.2*
Palmitoleic acid^{ns} (16:1)	7.8 ± 2.1	*4.0*	7.8 ± 2.2	*4.1*	3.1 ± 1.1	1.6	5.5 ± 1.8	*2.9*	9.4 ± 2.9	*4.7*
Stearic acid^{ns} (18:0)	30.3 ± 1.4	*15.6*	25.9 ± 1.6	*13.7*	29.5 ± 1.6	14.9	28.2 ± 1.7	*15.0*	28.2 ± 1.8	*14.2*
Oleic acid (18:1)	27.7 ± 1.6^{ab}	*14.2*	24.3 ± 1.7^{bc}	*12.8*	27.3 ± 1.2^{ab}	13.8	22.9 ± 1.0^{c}	*12.2*	29.6 ± 1.1^{a}	*14.9*
Linoleic acid^{ns} (18:2 n-6)	96.3 ± 5.6	*49.5*	99.5 ± 5.5	*52.5*	102.9 ± 2.6	51.9	103.2 ± 4.8	*54.9*	99.1 ± 3.1	*49.9*
Linoleic acid (18:3 n-3)	6.2 ± 0.3^{a}	*3.2*	4.5 ± 0.5^{b}	*2.4*	4.7 ± 0.6^{ab}	2.4	3.3 ± 0.7^{b}	*1.8*	4.2 ± 0.5^{b}	*2.1*
Total saturated fatty acids (SFA)^{ns}	56.5 ± 2.6	*29.1*	53.4 ± 3.2	*28.2*	60.2 ± 2.4	30.4	53.2 ± 3.4	*28.3*	56.4 ± 2.7	*28.4*
Total unsaturated fatty acids (SFA)^{ns}	137.9 ± 7.4	*70.9*	136.1 ± 6.9	*71.8*	138.0 ± 2.5	69.6	134.9 ± 5.0	*71.7*	142.3 ± 3.7	*71.6*
Total PUFA n-3 (or omega-3)	6.2 0.3^{a}	*3.2*	4.5 ± 0.5^{b}	*2.4*	4.7 ± 0.6^{ab}	2.4	3.3 ± 0.7^{b}	*1.8*	4.2 ± 0.5^{b}	*2.1*
Total PUFA n-6 (or omega-6)^{ns}	96.3 ± 5.6	*49.5*	99.5 ± 5.5	*52.5*	102.9 ± 2.6	51.9	103.2 ± 4.8	*54.9*	99.1 ± 3.1	*49.9*
n-6 to n-3 ratio	15.5^{ab}		22.1^{ab}		21.9^{ab}		31.3^{b}		23.6^{ab}	
UFA to SFA ratio^{ns}	2.4		2.5		2.3		2.5		2.5	

ns = not significantly different at P<0.05
Means within rows with different superscripts differ significantly at P>0.05

to total n-3 fatty acid ratios differed significantly (P<0.05) according to stables. Horses from the TTW stables had the worst (P<0.05) n-6 to n-3 ratio at 31.3 compared to the other stables which ranged from 15.5 to 23.6.

Plasma oleic acid of horses was also shown to differ significantly (P<0.05) according to the sampling locations. The TTW horses had different oleic acid level (P<0.05) compared to those from BTR, STC and RMP. The RMP horses also had the highest concentrations of plasma oleic acid with the TTW horses having the lowest amount among all horses sampled. The other plasma fatty acids were not significantly different (P>0.05) across sampling locations.

Hair coat scores

Based on the seven-point scale for skin scores (score 1 = worst score, score 7 = best score), the STC horses had the best hair coat score (median score = 7, P< 0.05). This was followed by horses from UPM (median score = 6), BTR (median score = 5), RMP (median score = 4) and finally horses from TTW (median score = 3). The hair coat scores for horses from the latter three stables (BTR, RMP and TTW) were not different from each other (Table 3).

Table 3: Mean hair coat scores by sampling locations

Locations	Median Score
STC 7^{a}	
UPM	6^{b}
BTR5^{bc}	
RMP	4^{bc}
TTW	3^{c}

Median values within column with different superscripts differ significantly at P<0.05

The results showed that there was a significant positive correlation between total n-3 fatty acids and hair coat scores (ρ = 0.686, P<0.01). This indicated that hair coat scores improved as plasma total n-3 fatty acid concentrations increase. There was also a significant inverse correlation between n-6: n-3 ratio and hair coat scores (ρ= -0.755, P<0.01). This meant that decreasing n-6: n-3 ratio was associated with better hair coat score in these horses.

DISCUSSION

Common horse feeds typically contain between 2 to 12 % fat (NRC, 1989). All the feeds surveyed in this

study had between 3 – 5% fat, which was within the normal dietary range for horses. All the feeds had different levels of fatty acids because they were collected from different locations and used different feed formulations. These feeds contained both linoleic acid and linolenic acid, reflecting the essentiality of these fatty acids in the equine diet. Feeds surveyed in this study contained high proportions of dietary linoleic acid but relatively lower linolenic acid. This is probably due to the high inclusion of grains in the equine diet, which is a rich source of n-6 fatty acids (Gurr *et al.,* 2002). The findings were also in line with the general agreement that the equine diet typically contains high amounts of linoleic acid (Lewis, 1996). However, feed storage conditions and handling would also determine the amount of n-6 and n-3 fatty acid levels over time. This is because the n-3 and n-6 fatty acids are more susceptible to peroxidation and rancidity compared to their saturated counterparts. Both hydrolytic and oxidative changes are involved in rancidity, which in turn are accelerated by heat, moisture, and light (Gunstone, 1996). Therefore, feeds that were not properly stored or handled in a cool, dry area with good air circulation might have contributed in part to the varying levels of n-3 and n-6 fatty acids observed in this study.

The plasma fatty acids levels, particularly the linoleic, oleic and linolenic acids, showed significant differences between locations. In general, plasma n-6 : n-3 fatty acid ratios seemed to be higher than 15, indicating a predominance of n-6 fatty acids in the plasma. This could be attributed to the different feed compositions that were fed to the horses at these locations and their associated environmental factors (Zembayashi and Nishimura, 1996), as dietary fatty acid profiles had long been known to influence the fatty acid composition of the plasma (Rajion *et al.,* 2001). However, changes in plasma oleic and palmitic acid levels are more difficult to interpret since they are also regulated by the action of tissue desaturases, and synthesised readily by the hepatocytes (Jenkins and Thies, 1997). Consequently, the levels of plasma fatty acids synthesised *de novo* is the result of complex interactions between the dietary lipids and the various intervening intrinsic and extrinsic factors within the animal itself (Chilliard, 1993).

Horses from different locations were also observed to have different hair coat scores. The skin of a healthy stabled horse should be elastic, smooth, clean and slightly warm. The coat should be fine, smooth, glossy and clean, giving the horse a sleek appearance (Pilliner and Davies, 1996). The skin can be affected by local problems but it is also the best indicator of the horse's general health and condition. The coat may become dry and dull if it is not lubricated by normal sebaceous secretions, or hidebound when the skin is tight and does not move freely over the underlying structure. This can be due to dehydration and lack of subcutaneous fat and is also seen in grass sickness and poorly nourished horses (Pilliner and Davies, 1996). Direct fatty acid supplementation played an important role in equine hair coat management apart from regular grooming, bathing and coat-clipping in horses (Pilliner and Davies, 1996). This is because n-3 and n-6 fatty acid deficiencies are known to predispose the skin to infection. These fatty acids are the constituents of epidermal sphingolipids and play an important role as barriers to exclude water and other molecules (Gurr *et al.,* 2002). Fatty acids such as gamma-linoleic acid (an n-6 fatty acid metabolite) had been shown to be able to modulate epidermal inflammation and hyperproliferation that might prevent poor hair coat and skin conditions (Ziboh, 1998). However, fluctuations in dietary EFA supplements could cause visible changes on the hair coat conditions (Goh *et al.,* 2002). Dominance of either n-3 or n-6 fatty acids determines the pro-inflammatory or anti-inflammatory tendencies of mammalian body. The n-3 fatty acids produce metabolites that are less inflammatory and this modulates hair growth promoting better skin health (Watkins and German, 1998). Horses sampled from the STC seemed have better hair coat scores compared to those from other locations. Horses included in this survey were healthy at the time of sampling, and had been groomed within five days before the hair coat scoring was performed. All of the horses had been on their present diet for at least two months, more than the minimum period required for dietary fatty acid supplementation to affect hair coat conditions (Rees *et al.,* 2001). Therefore, differences observed in the hair coat score could most probably be attributed to fatty acid-derived factors (Lewis, 1996). This is based on the fact that fatty acid molecules bonded to hair cuticle proteins are responsible for the hydrophobicity and luster of the hair coat (Rogers, 2004) apart from the factors that had been described in the earlier part of this paper.

Horses at the STC were fed diets with the highest unsaturated fatty acid: saturated fatty acid ratio. These horses also had the lowest plasma n-6: n-3 fatty acid ratio in their plasma and the best hair coat score. Conversely, horses from TTW had the lowest hair coat score in the study but the highest plasma n-6: n-3 fatty acids ratio. The relationship between n-6:n-3 ratio and hair coat conditions were further reaffirmed by the fact that the both are significantly correlated with each other ($\rho = -0.755$, $P<0.01$). However, only the plasma n-3 fatty acids were correlated with the hair coat score ($\rho = 0.686$, $P<0.01$) but not the n-6 fatty acids. This further illustrated the importance of n-3 fatty acids in the maintenance of the equine hair coat conditions in the tropical environment, especially since n-3 fatty acids are also known to protect the skin and hair coat from damage due to ultra violet radiation (Boelsma *et al.,* 2001).

The plasma n-6:n-3 fatty acid ratios are important to the hair coat conditions mainly because the n-6 and n-3 fatty acid families share the same enzymatic pathways in the animal body (Lee, 1993). The n-6 fatty acids are precursors for various inflammatory reactions while n-3 fatty acids help to reduce inflammatory response. The

likelihood of these fatty acids being metabolised to their respective anti-inflammatory or pro-inflammatory agents depended on the ratio of n-6: n-3 fatty acids in the body. More n-6 fatty acids are metabolised when the n-6: n-3 fatty acid ratio is high and vice versa. Increasing plasma n-6:n-3 fatty acid ratio has been associated with increased production of pro-inflammatory agent (Vaughn *et al.,* 1994). Therefore intake of n-6 and n-3 fatty acids must be regulated at a suitable ratio to avoid possible ill effects in the long run (Simopoulos, 1999). Miller *et al.,* (1991) devised a measure of the overall potential of dietary oils to exert local anti-inflammatory effect in guinea pigs. Their data demonstrated that dietary oils incorporated with n-3 and n-6 fatty acids influenced the distribution of polyunsaturated fatty acids in epidermal phospholipids. These eventually contributed to the ameliorative effect of oils on chronic inflammatory skin disorders. Although the n-3 and n-6 fatty acids can be used to correct skin and hair coat disorders, their excessive use had been known to trigger adverse effects (Harvey, 1993). Therefore, a balanced n-6 and n-3 fatty acid nutrition is important for equine skin health and hair coat conditions. There is currently no official recommendation of n-6 to n-3 fatty acid ratios for horses. However, it is recommended that n-6 and n-3 fatty acids be supplemented at the ratio of between 3:1 (n-6:n-3) and 5:1 (n-6:n-3) to achieve optimal effect on the equine hair coat.

CONCLUSIONS

In conclusion, this study showed that horse plasma fatty acids levels, particularly the linolenic acid, were different among horses from different locations. This was influenced by dietary fatty acids levels. The difference in plasma fatty acid levels, particularly the n-6 and n-3 fatty acids contributed to different hair coat conditions in these horses. Hair coat condition was found to correlate significantly with the total n-3 fatty acids and the n-6: n-3 fatty acid ratios in the plasma. High plasma n-3 fatty acid content and low n-6: n-3 ratio were associated with a better hair coat score in these horses.

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