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Carbohydrate And Glycogen Estimation in Two Insect Parasitic Nematodes Parasitizing The Gut of *Periplaneta Americana* at Meerut

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Abstract

During course of study of insect parasitic nematodes of Meerut region, the author extracted *Hammerschmiditiella diesingi* and *Schwenkiella oreintalis* from the gut of *Periplaneta Americana*. Quantitative estimation of carbohydrates and glycogen of female specimens of both these parasites was performed by the author. The amount of total carbohydrate appears to be very high in both the parasites. It was observed that total carbohydrate and glycogen content was more in *S. oreintalis* as compared to *H. diesingi*. While uptake of glucose was higher in *S. orientalis* as compared to *H. diesingi* but *H. diesingi* showed higher uptake of Sucrose. After six hours of incubation in various sugars, the percent depletion of endogenous glycogen content was observed to be different in different sugars.

Key words: *Hammerschmiditiella diesingi, Schwenkiella oreintalis, Quantitative estimation, Carbohydrate, Glycogen*

Introduction

As reported by various workers, insect parasitic nematodes can be employed as an agent of biological control to reduce the pest population without disturbing the ecology (Barron, 1981; Swarup and Gokte, 1986). During the course of study of insect parasitic nematodes of western U.P., the author came across two nematode species *viz.*, *Hammerschmiditiella diesingi* (Chitwood, 1932) and *Schwenkiella orientalis* (Singh and Agarwal, 1997), inhabiting the gut of *Periplaneta Americana*. Present communication deals with the biochemical composition of *H. diesingi* (Chit wood, 1932) and *S. orientalis* (Singh and Agarwal, 1997).

Materials and Methods

Biochemical studies of the nematodes were made as follows-Total sugar content of the parasites was determined by Anthrone method given by Yemm and Willis (1954). The alkali soluble glycogen was extracted ad estimated by the Anthrone method of Roe and Dailey (1966).

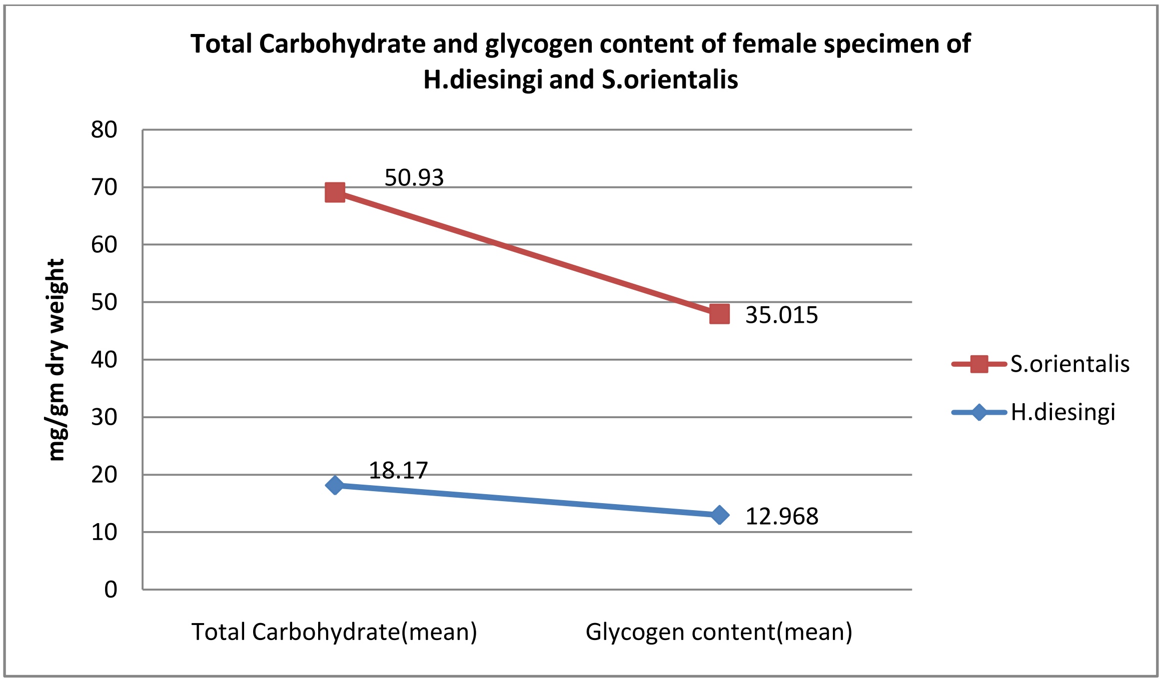
Observation

Results of quantitative estimation of various biochemical constituent of female specimen of *Hammerschmidtiella diesingi* Chit wood, 1932 and *Schwenkiella orientalis* Singh and Agarwal, 1997 are shown by Graphs.

Carbohydrate

The amount of carbohydrate appears to be very high in general in both the parasites Viz: *H. diesingi* and *S. orientalis*. Further the amount of carbohydrate appears to be more in *S. orientalis* as compared to *H. diesingi*. In case of former it amount to be 50.93 mg/gm in comparison to later which is 18.17 mg/gm.

On the other hand amount of glycogen also exhibits the same pattern ie. Higher in *S. orientalis* (35.015 mg/gm) and lower in *H. diesingi* (12.968 mg/gm)



Uptake of various Sugars

During the course of study, female live worms of both the genera were incubated in three different sugar solutions having equal concentrations. During the six hours of incubation, the utilization of glucose was found to be highest in *S.orientalis* but in case of *H.diesingi* utilitazation of sucrose was found to be highest (Table -1). Whereas *S. orientalis* utilize more glucose in comparison to *H diesingi*. During the course of this study it was observed that these worms prefer specific type of sugar, apart from their ability to utilize endogenous source of carbohydrates. The utilization of various sugars are observed in the following order-

Hammerschmidtiella diesingi **Chitwood,1932**

Sucrose>Glucose >Lactose

Schwenkiella orientalis **Singh and Agarwal,1997**

Glucose >Lactose> Sucrose

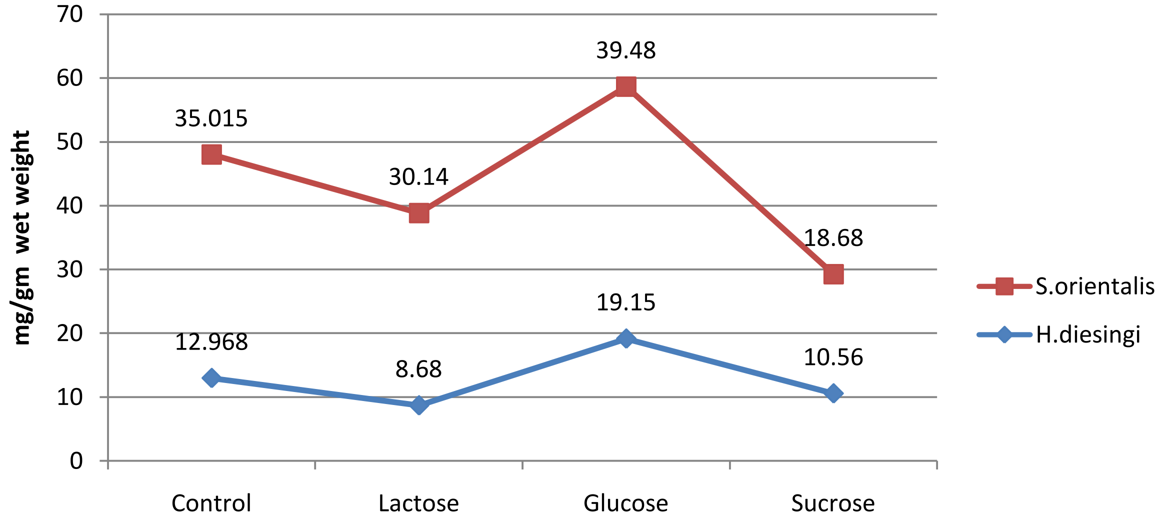
Table–1: Uptake of various sugars (in mg/gm of fresh tissue weight) in female specimens of *H. diesingi* Chitwood, 1932 and *S. orientalis* Singh and Agarwal, 1997

		Sugars	
	Lactose	**Glucose**	**Sucrose**
H. diesingi	31.38	45.48	60.24
S. orientalis	80.35	285.41	72.1

This indicates that worms have a preferential uptake of sugars. In case of *H. diesingi* the maximum absorption is of sucrose followed by glucose and lactose. But in case of *S. orientalis* it is glucose which shows the maximum absorption followed by lactose and sucrose. The most note worthy thing is that though the sugars were provided in same concentration but the uptake is different from each other (Table–1). From the Table-1 it is also evident that uptake of lactose is about 2.5 times higher in *S. orientalis*, absorption of glucose is about six times higher in *S. orientalis* and absorption of sucrose is about 1.2 times higher in *S. orientalis* when it is compared with *H. diesingi*.

Table2: Endogenous glycogen contents (mg/gm wet weight) of female specimens of *H. diesingi* Chitwood, 1932 and *S.orientalis* Singh and Agarwal, 1997 after six hours of incubation in various sugars

	Control	**Lactose**	**Glucose**	**Sucrose**
H. diesingi	12.968	8.68	19.15	10.56
S. orientalis	35.015	30.14	39.48	18.68



Endogenous glycogen content of female specimens after 6 hours of incubation in various sugars

A depletion is also observed in the endogenous glycogen content of these worms, after incubation of six hours in different sugars .The percent depletion is observed to be different in different sugars (Table -2 & 3). The maximum depletion is observed in case of *S. orientalis* when it was incubated in glucose solution followed by sucrose and lactose. However, in case of *H. diesingi* maximum depletion is noticed in incubation in lactose followed by glucose and sucrose.

Table -3: Percent depletion in glycogen content of female specimens of *H. diesingi* Chitwood, 1932 and *S. orientalis* Singh and Agarwal , 1997 after six hours of incubation in various sugars

	Lactose	**Glucose**	**Sucrose**
H. diesingi	39.48	34.15	20.76
S. orientalis	23.15	60.16	59.56

Discussion

Carbohydrates

It has been known for more than hundred years that parasitic worms contain polysaccharide (Bernard, 1859; Foster, 1865). All the worms living in anaerobic or in semi anaerobic habitats, for example in intestine or in bile ducts and having no specific means of securing oxygen, utilize carbohydrates primarily. Because their immediately oxidized carbon atoms are suited ideally for oxidative processes. While the parasites living in oxygen rich surroundings, for example blood, drive most of their energy from the oxidation of fats and proteins but carbohydrate metabolism never predominates in such worms (Von Brand, 1973). Unfortunately, no definite reason for this specialization has been recognized as yet, the main energy source for parasitic nematode is the carbohydrate (Barrett, 1976).

Total carbohydrate

The total carbohydrate in *H. diesingi* is estimated to be 0.18 in percent of their fresh weight while in *S.orientalis* the value is 0.509. It shows higher carbohydrate content in *S. orientalis* as compared to *H. diesingi*. Earlier following workers have contributed to our knowledge on the carbohydrate content in the nematode, parasitic in certain vertebrates like (Faure- Fremiet,1913) reported presence of 0.15% sugar in the body fluid of *Parascaris* species. Castro and Fairbairn (1969) reported the presence of 21.10% carbohydrate content in the larva of *Trichinella spiralis*. Naidu, (1980) estimated the amount of carbohydrate in *Bunostomum trigonocephalum* and reported 1.37% carbohydrate in males and 2.2% carbohydrate in females. *Rathaur et.al.*, (1980) reported 1.48% carbohydrate in adult female of *Setaria cervi* and 8.97% in the microfilaria of the same species. Rogers (1959) and Crompton et. al. (1965) demonstrated that their occur steep oxygen gradient between the intestinal mucosa and intestinal lumen. Further Von Brand, (1973) observed that all parasitic helminths are facultative aerobes and are capable of utilizing oxygen as and when available, which was further confirmed by Murlidhar and Rao (1981) in case of *Ascaridia galli.*

In case of *H. diesingi* and *S. orientalis* it can be presumed that chiefly they are anaerobes since oxygen availability is very less because of being intestinal parasite. It has been earlier observed that parasite living in oxygen deficient habitat or in an environment with periodic oxygen deficiencies as in case of intestine, usually have rich carbohydrate reserve for their survival under adverse environmental conditions (Von Brand, 1973).

Presence of higher carbohydrate content in *S.orientalis* in comparison to *H. diesingi* could be due to the fact that this parasite resides on the outer surface of intestinal folds of the host, it is likely that the food supply may be affected many times. This corrobates the findings of Rathaur et. al. (1980), Who also reported similar findings in *Setaria cervi.* The lower amount of carbohydrate in *H. diesingi* could also be due to the fact that this parasite might be inhabiting a place in the intestine, having poor flow of digested food material, so there is no need to store carbohydrate as reserve food material. Almost similar findings has been documented by Srivastava et.al. (1970) in *Litomosoides carinii.*

More carbohydrate in *S. orientalis* as compared to *H. diesingi* has been studied. This difference in carbohydrate content in *S. orientalis* and *H. diesingi* is also because of the fact that rectal region in which the *H. diesingi* is found quieter, where it gets the carbohydrate only from surrounding tissues of the host, which is already deficient in carbohydrate reserves. Besides this, since it inhabit a part of alimentary canal , it does not have to effort to maintain its position in the alimentary canal, as it is quite .Therefore , it has got less carbohydrate in comparison to *S. orientalis* inhabiting very active region of alimentary canal having rich supply of carbohydrate. Besides this, in this intestine always there is some activity going on, like movement of the bowl, peristalsis. Thus the parasite has to make effort to sustain in that part of the alimentary canal .Thus they have to have higher carbohydrate reserve as source of energy.

Glycogen

The most important storage polysaccharide of animal parasitic nematode is glycogen (Von Brand, 1952). The glycogen content of *H. diesingi* under normal condition is 12.968 mg/gm and in

S. orientalis is 35.015 mg/gm of their fresh weight which indicates that both these worms have significantly high amount of glycogen.

Different workers have worked out the glycogen content of various nematode parasites of vertebrate inhabiting different host like -

Ascaris lumbricoides having glycogen content 3.3-3.87% (Weinland, 1901; Flury, 1912; Smorodincev and Bebesin, 1936; Cavier and Savel,1951); 2.11-3.8% in *Parascaris equorum* ; 2.0- 2.4% in *Filaria equana*; 3.5% in *Strongylus vulgaris* (toryu, 1933) ; 1.6% in *Ancylostoma caninum* (Von Brand and Otto , 1938; Fernando and Wong , 1964; Clark , 1969); 0.8% in *Litomosoides carinii* (Bueding, 1949); 0.2% in *Dipetalonema gracilis*; 1.9% in *Dirofillario immitis* (Von Brand, 1950); 2.7% in *Heterakis gallinae* (Glocklin and Fairbairn, 1952); 2.3% in *Trichuris vulpis* (Bueding et. al., 1961); 2.3% in *Setaria cervi* (Pandya, 1961; Rathaur et.al. 1980); 1.7% in *Dirofillaria immitis* (Von and Brand, 1963); 0.088% in *Nippostrongylus brasilliensis* (Roberts and Fairbairn, 1965; Wilson, 1965) ; 0.9% in *Angiostrongylus cantonensis* (Yanagisawa and Von Brand , 1965); 9.3-11.7% in *Strongyluris brevicaudata* (Umerzurika and Anye, 1978); 3.95-4.87% in *Bunostomum trigonocephalum* (Gupta and Trivedi, 1984); 1.10- 1.39% in female of *Oesophagostomum columbianum* (Premvati and Chopra, 1979) and 1.22- 1.46 % in female of *Haemonchus contortus* (Premvati and Chopra, 1979).

According to Halton, (1967) and Von Brand, (1973) the parasite which inhabit an environment having high oxygen content possess low glycogen where as those parasite which inhabit anaerobic condition posses high glycogen. Presence of higher glycogen content in both the nematodes under study is basically due to their presence in an environment having low oxygen content. Which has also been observed by workers like Reid, (1944); Srivastava et.al., (1970); Premvati and Chopra, (1979) and Murlidhar and Rao, (1981).

It is difficult for the author to corrobate the presence of higher glycogen content in the parasite inhabiting intestine and the other inhabiting rectum. The specimen of *S. orientalis* obtained from intestine have almost three times more glycogen as compared to specimens of *H. diesingi* obtained from rectum of the same host as practically no attempted made by earlier workers on this line . However, in my opinion, it could be due to following reasons:

1. Somehow oxygen supply is more in the rectum as compared to intestine.

2. It could be due to the reason that parasite inhabiting rectum fail to get enough morsels to convert them into reserve food material.

3. This could also be due to the presence of symbiotic protozoans in the rectum of the same host.

Uptake of glucose

It is an established fact that parasite utilize exogenous glucose when made available to it (Barrett, 1976). After six hours of incubation the glucose utilization by *H. diesingi* is 45.48 mg/gm fresh weight whereas in *S. orientalis* it reaches to 285.41 mg/gm fresh weight. The utilization of exogenous glucose has been studied by number of workers like Bueding, (1949), Bueding and Oliver -Gonzalez (1950), Von Brand et. al., (1963) . Von Brand (1973) made a comprehensive review of earlier studies on this line and reported that it varies from species to species. Cavier and Savel, (1951) reported that the cuticle of *Ascaris lumbricoides* is impermeable to soluble sugars whereas cuticular absorption of glucose has been reported in *Ascaridia galii* by Weatherly et. al.,(1963). This indicates that

1. Major portion of glucose is absorbed through established openings and

2. Remaining portion is absorbed through the cuticle. Author also agrees with Weatherly et.al (1963).

Uptake of various sugars

During the course of study, it was observed that maximum absorption of sucrose is noticed in case of *H. diesingi* whereas maximum absorption of glucose is noticed in *S. orientalis*. This proves that parasites can make use of other sugars than glucose which has also been reported by Cavier and Savel, (1951). Besides this, it was also noticed that the rate of uptake of sugar is many times higher in case of *S. orientalis* as compared to *H. diesingi*. It is difficult for the author to corrobate her findings with earlier workers. As practically, no attention has been paid by workers in past to study the uptake of various sugars. However, in my opinion *S. orientalis* is a parasite inhabiting intestine, thus it requires more energy in comparison to *H. diesingi* inhabiting comparatively calm environment ie. Rectum.

Glycogen content after incubation in various sugars

During the course of study, a depletion of glycogen content was observed in *H. diesingi* and *S. orientalis* after six hours of incubation . More over it was also noticed that the percent depletion is different in different sugars. It is difficult for the author to correlate her findings as practically no attempt has been made by helminthologist in past to study this aspect of nematode biology.

In case of *H .diesingi* maximum depletion was noticed in case of lactose (39.48%) followed by glucose (34.15%), followed by sucrose (20.76%). However in case of absorption of these sugars this nematode exhibit reverse trend. Maximum absorption was noticed in case of sucrose followed by glucose and finally the lactose.

Moreover, in *S. orientalis* the maximum depletion was noticed in case of glucose (60.16%) followed by sucrose and lactose (59.56 and 23.15 % respectively), which is contradictory. In my opinion this could be due to the fact that *S. orientalis* is an active parasite and during in vitro cultivation in different sugar solutions the parasite rather than absorbing glucose, was also busy in finding better hide-outs. Thus, the energy depots present in the body of parasite get utilized rapidly.

Reference

Barrett, J. 1976. Bioenergetics in Helminths, In : Biochemistry of Parasites and Host Parasite relationships. (H. Van. Den. Bosche ed.) Falsevier / North Holland Boimedical Press, Amserdam.

Barron, G. L. 1981. Predators and parasites of microscopic animals.
In biology of conidial Fungi, Volume 2 (edited by Cole, G. T. Rendrick, B)

Academic Press 167 - 200.

Bernard, C. 1859. De La Matiere glycogene chez les animaux depousves de foie. Comp. Rend. Soc. Biol. Ser., 3: 53 - 55

Bueding, E. 1949. Studies on the metaboliem of the filarid worm Litomosoides carinii. J. Exp. Med. 89: 107 - 136.

Bueding, E.; Kmetec, E. Swartzweider, C.; Abadie, S.; and Saz, H. J. 1961. Biochemical effects of dithiazanine on the canine whip woem, Trichuris,vulpis. Biochem. Pharmac., 5: 311 - 322.

Bueding, E. And Oliver- Gonzalez, J. 1950. Aerobic and anaerobic production of lactic acid by filarial worm, Dracunculus insignis. Brit. J. Pharmac. Chemather., 5: 62 - 64.

Castro, G. A. And Fairbairn, D. 1969. Carbohydrates and Lipids in Trichinella spiralis larval and thrie utilization in vitro. J.Parasitol., 55: 51 - 58.

Cavier, R . And Savel, J. 1951. Chemical composition of Ascaris lumbricoides from swine. Bull. Soc. Chim. Biol., 33: 447 - 454.

Chitwood, B. G. 1932. A synopsis of nematodes parasitic in insects of the family Blattidae. Z. Parasitenk, 5: 14-50.

Clark, F. E. 1969. Food reserved and changes in chemical composition with age in third stage larvae. Exp. Parasitol., 24:1-8

Crompton, D. W. T. ; Shrimption, D. H. and Silver, J. A. 1965. Measurement of the oxygen tension in the lumen of the small intestine of the domestic duck. J. Exp. Biol., 43: 473 - 478.

Davies, R. G. 1977. Imms general text book of entomology,Chapman and Hall, New York.

Faure- Fremiet, E. 1913. Arch. Anat. Microsc. Morphol, Exp. 15: 435 - 457. (Not consulted in original cited from Basir, 1940.)

Fernando, M. A. and Wong, H. A. 1964. Metabolism of Hook worms II. Glucose metabolism and glycogen synthesis in adult female (Ancylostoma caninum). Exp. Parasitol., 15: 1284 - 1292.

Flury, F. 1912. Zur chomie Und Toxikologic der Ascariden. Archiv. Fur Experimentella Pathologie Und Pharmakologic, 67: 275 - 392.

Foster, M. 1865. On the existence of glycogen in the tissues of certain entozoa. Proc. Roy. Soc. London, 14: 543 - 546.

Gupta, S. P. and Trivedi, K. K. 1984. Biochemical studies on in-vitro variation of glycogen content in sheep nematode, Bunostomum trigonocephalum Rudolphi, 1808. Pakistan Journal of Zoology, 16 : 83 - 90.

Halton, D. W. 1967. Comp. Biochem. Physiol., 23 : 113 -12 . (cited from Von Brand, 1973).

Imms, A.D. 1960. A general text book of entomology, Methuen, London

Lee, D. L. And Atkinson, H. J. 1976. Physiology of nematodes. Macmillan Press, London.

Murlidhar, A. and Rao, L. N. 1981. Metabolic differenciation of male and female Ascaridia galli: Glycogen Pyruvate and lactate contents. Proc. Ind. Acad. Parasitol., 2 :7 - 9.

Naidu, T. S. V. 1980. Gross chemical composition of Bunostomum trigonocephalum (Rud., 1808) Railliet,1902. Rivista di Parasitologia, 41: 207 - 208.

Pandya, G. T. 1961. Physiological studies on nematodes, Setaria cervi I. Carbohydrate metabolism. Z. Parasitkde, 20: 466 - 469.

Premvati, G. and Chopra, A. K. 1979. In vitro variation of glycogen content in three sheep nematodes. Parasitology, 78: 355 - 359.

Rathaur, S. ; Anwar, N. and Ghatak, S. 1980. Biochemical composition of microfilaria and adult stages of Setaria cervi. Zeitschrift Fur Parasitenkunde, 62: 85 - 93.

Reid, W. M. 1944. The effect of host starvation on worms elimination and glycogen depletion with the nematode Ascaridia galli. J. Parasitol. 30: 12.

Roberts, L. S. and Fairbairn, D. 1965. Metabolic studies on adult Noppostrongylus brasilensis (NematodA: Trichostrongyleidae). J. Parasitol., 51:129 - 138.

Rogers, W. P. 1949. On the relative importance of aerobic metabolism in small nematode parasites of alimentary canal-II. The utilization of oxygen at low partial pressure by small nematode parasites of alimentary tract. Aust. J. Sci. Res. Section B_2: 166 - 174.

Singh, H. S. and Agarwal, A. 1997. On a new nematode, Schwenkiella orientalis new sp., from Periplaneta americana Linn. U.P.J. of Zoology, 17: 123 - 126

Smorodincev, I. A. and Bebesin, K. W. 1936. La teneur englycogene des adcaridis Dolk. Akad. Nauk. SSSR (NS), 2: 189 - 191

Srivastava, V. M. L.; Ghatak, S. and Krishna Murti, C. R. 1970. Ascaridia galli Lactic acid production, Glycogen content, Glycolytic enzymes and properties of purified aldolase enolase and glucose 6 phosphate dehydrogenase, Parasitology, 60: 157 - 180.

Swarup, G. And Gokte, N. 1986. Biological control, Plant parasitic nematodes of India Ed: Swarup and Das Gupta, IARI Publication 476 - 489 pp.

Toryu, Y. 1933. Contribution to the physiology of the Ascaris I. Glycogen content of Ascaris. Ascaris megalocephala. Cloq. Sci. Rep. Tohoku Univ., 8: 65 - 74.

Umezurika, G. M. and Anye, D. A. 1978. Studies on the energy metabolism in the endoparasite Strongyloides brevicaudata (Nematoda) Comp. Biochem. Physiol., 59: 147 -151.

Von Brand, T. 1952. Chemical physiology of endoparasitic animals. Academic Press, New York.

Von brand, T. 1973. Biochemistry of Parasites. Academic Press, New York and London.

Von Brand, T. ; Bowman, I. B. R. ; Weinstein, P. P. and Sawyer, T. K. 1963. Observation on the metabolism of Dirofilaria uniformis. Exp. Parasitol.,13 : 128 - 133.

Von brand, T. and Otto, G. F. 1938. Some species of carbohydrate metabolism of the Hook worm, Ancylostoma caninum. Americana J. Hug.n. 27: 683-689.*

Weatherly, N. F. ; Hansen, M. F. And Moser, H. C. 1963. Exp. Parasitol., 14: 37 - 48 (cited by Von brand, 1973).

Weinland, E. 1901. Z. Biol., 42:55-90 (as cited by Von brand, 1973).

Wilson, P.A. G. 1965. Changes in lipid and nitrogen content of Nippostrongylus brasilensis infective larvae aged at constant temperature. Exp. Parasit., 16:190 - 194.

Yanagisawa, Y. and Von Brand, T. 1965. Carbohydrate metabolism in Angiostrongylus cantonensis. J. Parasitol., 51: 418 - 423.