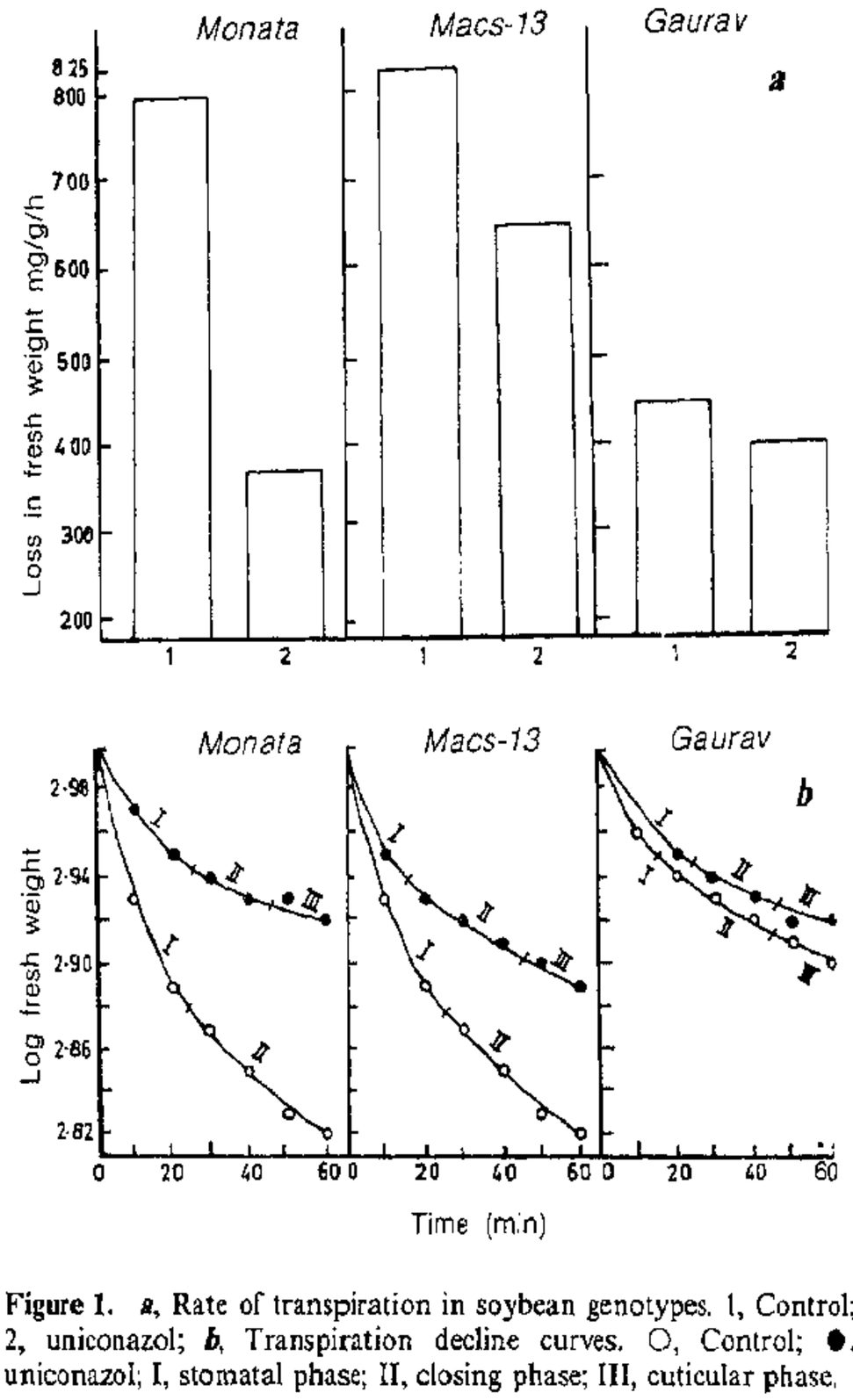

Figure 1. *a*, Rate of transpiration in soybean genotypes. 1, Control; 2, uniconazol; *b*, Transpiration decline curves. ○, Control; ●, uniconazol; I, stomatal phase; II, closing phase; III, cuticular phase.

ration rates in all the three genotypes of soybean compared to their respective untreated controls. The decrease in rates was of the order of 53, 21 and 10% in *monata*, *macs-13* and *gaurav* respectively. An analysis of the transpiration decline curves shows predominance of stomatal and closing phases in control of *monata* and *macs-13* whereas the cuticular phase appeared in all the treated plants of all the genotypes and control of *gaurav* (Figure 1*b*). It is apparent that uniconazol reduced the transpiration rate by decreasing the magnitude of stomatal transpiration through partial stomatal closure and hence exhibited antitranspirant-like activity. Recently Santakumari and Fletcher[7] reported that triazoles partially close stomata in isolated epidermal strips of *Commelina benghalensis* L.

Since most research has been aimed at controlling stomatal aperture with the ultimate objective of maintaining a favourable internal water balance[6], the use of uniconazol may prove to be useful for enabling plants to withstand drought condition by preventing water loss, and keeping a high internal water balance. This view is supported by the fact that a related triazole-paclobutrazol-treated plants showed high water potential compared to control[8]. Further, it is noted that the use of PMA as an antitranspirant does not seem to be safe, since it is poisonous on many enzymatic systems[9] and drastically affects the chlorophyll content[6]. Hence it is apparent that the use of plant-growth regulators like uniconazol as antitranspirant holds great promise and make plants suitable for dryland cultivation without any adverse effects on the plants.

1. Bora, K. K., Ph.D. thesis, University of Jodhpur, 1981.
2. Bora, K. K. and Mathur, Sudha, *Comp. Physiol. Ecol.*, 1989, **14**, 159.
3. Bora, K. K., Sankhla, D. and Sankhla, N., *Proc. Natl. Sem. Plant Physiol.*, 1984, p. 110.
4. Bora, K. K., *Ann. Arid Zone*, 1988, **27**, 301.
5. Bora, K. K. and Sharma, D. D., *Biochem. Physiol. Pflanzen*, 1988, (in press).
6. Waisel, Y., Borger, G. A. and Kozlowski, T. T., *Plant Physiol.*, 1969, **44**, 685.
7. Santakumari, M. and Fletcher, R. A., *Physiol. Plant.*, 1987, **71**, 95.
8. Wieland, W. F. and Wample, R. L., *Sci. Hortic.*, 1985, **26**, 129.
9. Zelitch, I., *Conn. Agric. Exp. Stn. Bull.*, 1963, **644**, 18.

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Amino acid carbohydrate transformations in adult *Setaria cervi* females

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Adult *Setaria cervi* females were able to incorporate exogenous U-^{14}C-glucose into proteins and U-^{14}C-chlorella protein hydrolysate into glycogen. Enzymes involved in this transformation were detected in crude homogenates of the worms. The activities of L-glutamate oxaloacetate transaminase (EC 2.6.1.1), L-glutamate pyruvate transaminase (EC 2.6.1.2) and L-glutamate dehydrogenase (EC 1.4.1.3) were around 8660 ± 114; 12600 ± 391 and 0.27–0.90 nmol/min/mg protein respectively.

VERY few reports are available on the relationship between amino acids and carbohydrates in filarial group of nematodes[1,2]. We had earlier described a cell-free system for protein biosynthesis in a bovine filarial nematode *Setaria cervi* adult females[3]. This paper reports that adult *S. cervi* females could form glycogen from a mixture of amino acids and proteins from glucose.

The methods for collection, incubation and preparation for enzymes of *S. cervi* adults have been described elsewhere[4]. The adult *S. cervi* females were collected

from freshly slaughtered naturally infected water buffaloes (*Bubalus bubalis* Linn.) at a local abbatoir soon after decapitation, and brought to the laboratory in physiological saline. For incubation studies, the actively motile worms (100–150 mg wet weight) were placed in 10.0 ml of Krebs Ringer bicarbonate medium (KRB) containing either 37.0 kBq U-^{14}C-glucose (5.55 mM) or U-^{14}C-chlorella protein hydrolysate (1.0 mg/ml). After 1 h of incubation the worms were removed, washed thoroughly with fresh chilled KRB containing the respective cold carrier and homogenized in ice-cold glass distilled water. Macromolecular fractions were separated[5] and the amount of radioactivity was measured by Packard liquid scintillation counter[6]. For enzyme studies, the actively motile worms were homogenized in 10 volumes of 150 mM KCl (w/v). Homogenates were centrifuged at 1,000 *g* for 15 min at 4°C; supernatants were analysed for L-glutamate oxaloacetate transaminase (EC 2.6.1.1), L-glutamate pyruvate transaminase (EC 2.6.1.2) and L-glutamate dehydrogenase (EC 1.4.1.3) activities according to Reitman and Frankel[7], and Langer and Bernhardt[8].

Figures 1 and 2 show the amount of radioactivity recovered in different fractions of *S. cervi* adult females after incubation with U-^{14}C-glucose and U-^{14}C-chlorella protein hydrolysate. Nearly 2–3% of the total radioactivity from U-^{14}C-glucose was found in the protein fraction and about 3–5% of the total radioactivity from U-^{14}C-chlorella protein hydrolysate was associated with the glycogen fraction after 1 h of incubation.

Tables 1 and 2 show the activities of L-glutamate oxaloacetate transaminase, L-glutamate pyruvate transaminase and L-glutamate dehydrogenase in cell-free extracts of *S. cervi* adult females. Of the two transaminases, glutamate pyruvate transaminase was more active than glutamate oxaloacetate transaminase. The specific activities of the system which deaminate glutamate with either NAD^+ or NADP as cofactor show that the reaction proceeds more rapidly when NAD^+ was used. Glutamate formation by the L-glutamate dehydrogenase proceeds essentially at the same rate regardless of the cofactor (either NADH or NADPH) employed.

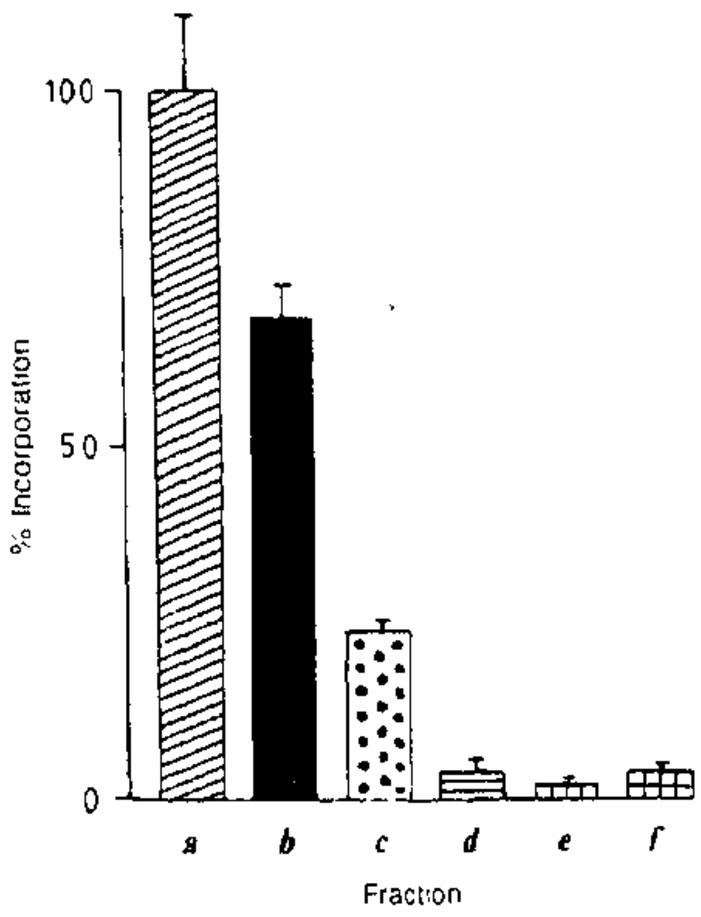

Figure 1. Distribution of radioactivity after incubating adult *Setaria cervi* females in Krebs Ringer bicarbonate medium containing U-^{14}C-glucose (5.55 mM). ***a***, Whole homogenate; ***b***, cold trichloroacetic acid soluble fraction; ***c***, glycogen fraction; ***d***, trichloroacetic acid precipitable protein fraction; ***e***, lipids; ***f***, nucleic acid fraction.

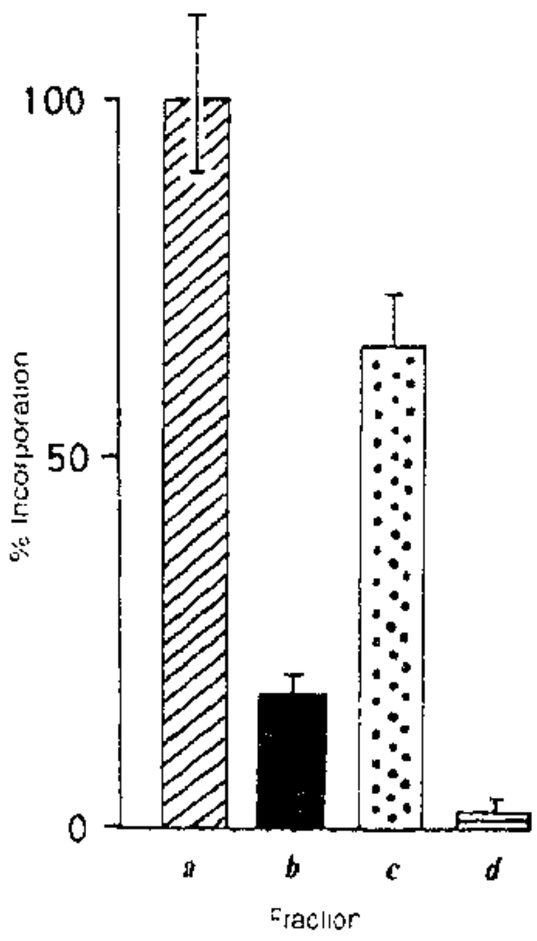

Figure 2. Distribution of radioactivity after incubating adult *Setaria cervi* females in Krebs Ringer bicarbonate medium containing U-^{14}C-chlorella protein hydrolysate (1.0 mg/ml). ***a***, Whole homogenate; ***b***, trichloroacetic acid precipitable protein fraction; ***c***, trichloroacetic acid soluble fraction; ***d***, glycogen fraction.

Table 1. Activity of L-glutamate oxaloacetate transaminase, and L-glutamate pyruvate transaminase in cell-free extracts of adult *Setaria cervi* females.

Enzyme	Activity*
L-Glutamine oxaloacetate transaminase	8660 ± 114
L-Glutamine pyruvate transaminase	12600 ± 391

*Values are expressed as nmol/min/mg protein ± SE, based on triplicate samples of three separate batches.

Table 2. Activity of L-glutamate dehydrogenase in cell-free extracts of adult *Setaria cervi* females towards various substrates and cofactors.

Substrates used	Cofactor employed	Activity*
L-Glutamine	NAD	0.90 ± 0.12
-do-	NADP	0.27 ± 0.09
α-Ketoglutarate, and NH_4^+ ions	NADH	0.76 ± 0.21
-do-	NADPH	0.61 ± 0.11

*Values are expressed as nmol/min/mg protein ± SE, based on triplicate samples of three separate batches.

The simplest way by which the carbon atoms derived from glucose may be incorporated into amino acids and finally into the proteins occurs by reductive amination reaction which are catalysed by glutamate dehydrogenase[9]. A comparison of amination and deamination reactions catalysed by L-glutamate dehydrogenase indicates that the amination of α-keto acids is more active than the deamination reactions (table 2). Since α-keto acids are the common intermediates of glucose utilization in the filarial worms[1], it is not surprising that reductive amination of these amino acids is the basis for the appearance of radiocarbon of glucose in protein fractions like animal cells in these nematodes. Enzymes catalysing the transamination reactions between amino acids and α-keto acids are also present in *S. cervi* adult females which may also be another link between carbohydrate and amino acid metabolism. A reverse tricarboxylic acid cycle has been reported in many filarial species[10-12]. On this basis it can be proposed that the incorporation of carbon atoms of chlorella protein hydrolysate into glycogen fractions of *S. cervi* adult females is the result of deamination of amino acids of chlorella protein hydrolysate and also their entry into tricarboxylic acid cycle.

1. Von Brand, T., in *Biochemistry and Physiology of Endoparasites* (ed. T. Von Brand), Elsevier/North Holland, Amsterdam and London, 1979.
2. Barrett, J., *Helminthol. Abstr. Ser.*, 1983, **A52**, 1.
3. Srivastava, A. K. and Ghatak, S., *Med. Sci., Res.*, 1989, (submitted).
4. Srivastava, A. K., *Chemical structural and metabolic studies in filariasis*, Ph.D. thesis, University of Kanpur, Kanpur, 1980.
5. Rathaur, S., Anwar, N. and Ghatak, S., *Z. Parasitenkd.*, 1980, **62**, 85.
6. Srivastava, A. K. and Ghatak, S., *Curr. Sci.*, 1983, **52**, 303.
7. Reitman, S. and Frankel, S., *Am. J. Clin. Pathol.*, 1957, **28**, 56.
8. Langer, Jr. and Bernhardt, W., *J. Parasitol.*, 1972, **58**, 539.
9. Pollak, J. K. and Fairbairn, D., *Can. J. Biochem. Physiol.*, 1955, **33**, 307.
10. Barrett, J., Mendis, A. H. W. and Butterworth, P. E., *Int. J. Parasitol.*, 1986, **16**, 465.
11. Walter, R. D. and Albiez, E., *Trop. Med. Parasitol.*, 1986, **37**, 96.
12. Middleton, K. R. and Saz, H. J., *J. Parasitol.*, 1979, **65**, 1.

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Erythrocytie membrane changes in sheep infected with *Dictyocaulus filaria*

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Lambs infected orally with 2500 *Dictyocaulus filaria* larvae showed a significant fall in levels of erythrocyte membrane cholesterol, cholesterol: phospholipid ratio and acetylcholinesterase activity from fourth to tenth week ($P<0.05$ to $P<0.001$) post-infection. Plasma cholesterol levels in infected lambs were also significantly different from fifth week onwards from those in uninfected controls. These alterations set in one to two weeks prior to increase in osmotic fragility of erythrocytes and coincided with development of the parasite to adult stage in the infected host.

An increase in the osmotic fragility of erythrocytes has been reported in various diseases of livestock[1] and in toxic conditions[2]. Recently, increased osmotic fragility of sheep erythrocytes was reported in *Dictyocaulus filaria* infection[3]. This communication reports our preliminary findings on erythrocyte membrane changes *vis-a-vis* decreased resistance of sheep erythrocytes to osmotic lysis in *D. filaria* infection.

The study was carried out in *D. filaria* producer lambs maintained at this laboratory for vaccine production. Eight producer lambs were randomly selected and grouped into acute infection (5 lambs) and chronic infection (3 lambs) groups based on day of infection. The zero week post-infection (PI) observation for various parameters in the chronic infection group corresponds to 27th week PI. Each animal in these groups had received an infection dose of 150 *D. filaria* larvae per kg body weight. Three animals were also maintained as uninfected controls. The osmotic fragility of erythrocytes was determined as described earlier[3]. Standard methods were employed for the preparation of erythrocyte membranes[4], estimation of membrane cholesterol and phospholipids[5], and determination of acetylcholinesterase activity[6]. The study was carried out for ten weeks.

The lambs in acute stage of infection showed a decrease from fourth week onwards in membrane cholesterol (0.89 ± 0.13 to 1.31 ± 0.14 mg/ml packed cells), cholesterol to phospholipid ratio (0.61 ± 0.13 to 0.84 ± 0.12) and acetylcholinesterase activity (10.56 ± 1.75 to 16.36 ± 1.8 mol/min/RBC$\times10^{-22}$) compared to uninfected controls (cholesterol, 1.38 ± 0.09 to 1.45 ± 0.1; cholesterol to phospholipid ratio, 0.97 ± 0.09 to 1.00 ± 0.08; acetylcholinesterase activity, 18.88 ± 1.75 to 18.76 ± 1.95). However, group differences were significant from fourth or fifth week PI ($P<0.05$ to $P<0.001$.) Membrane phospholipids in the infected lambs remained unaffected during the course of infection. The changes