

METABOLIC STUDY OF CYCLE RICKSHAW PULLERS BEFORE AND AFTER EXERCISE

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ABSTRACT ■ Studies regarding metabolic pattern of cycle rickshaw pullers (RP) due to repeated strenuous exercise are scanty. This study aimed at assessing nutritional status of RPs and to evaluate the effects of repeated strenuous exercise on their metabolic status. Thirty five cycle rickshaw pullers and 21 control subjects were included. Rickshaw pullers were asked to pull the rickshaw in six cycles. Controls were allowed to work with a cycle ergometer. At the outset, 24 hour urinary creatinine, urinary sodium, potassium, blood Hb% value was estimated. Plasma glucose, serum urea, total protein, albumin, triglyceride, cholesterol, creatine kinase (CK), CK-MB isoenzyme, gamma glutamyl transpeptidase (GGT), pyruvate, lactate, free fatty acid, Na and K were estimated before and after strenuous exercise. There was significant increase in glucose, triglycerides, urea, CK, pyruvate and lactate in both groups after exercise. CK-MB showed significant increase only in controls. Similarly, post-exercise mean lactate was higher in controls. Lactate/pyruvate ratio, free fatty acid, serum potassium significantly decreased after exercise in both groups. Significantly low values of 24-h urinary Na and creatinine were observed in RPs than controls before exercise. Increase in lactate was far less than that of controls suggesting higher exercise tolerance. Lactate/pyruvate ratio less than 25 both in pre and post exercise state of both groups indicated that during exercise there was efficient gluconeogenesis. Low values of 24-h urinary Na and creatinine prior to exercise indicated that renal reabsorption capacity in RP were higher perhaps resulting from adaptive response to altered metabolic status.

Key words: Cycle Rickshaw pullers, Plasma Glucose, Triglyceride, Creatine kinase, Lactate

INTRODUCTION

The cycle rickshaw is a common mode of transport which is used as a cheap mean of conveyance for small distances. It is designed to carry two passengers along with their luggage and merchandise. In India, rickshaw pulling is a popular occupation of poor and low socio-economic class for earning basic need. About 0.86 million people is engaged

in this occupation out of which 0.26 million (30%) are in West Bengal (Vijayanunni 1991). Cycle rickshaw pullers (RP) work for long hours; there is no fixed time for their work. As supported by earlier observations (Astrand and Rodahl 1986, Pradhan et al 2004, Sen and Nag 1975), activities of pullers showed the workload as 'heavy' and 'very heavy' category which may influence their

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biochemical and haematological characteristics. Though the relative contributions of the various means of energy transfer differ markedly depending on the intensity and duration of the exercise and the fitness of the person, several studies have been done to explain the effect of exercise on intermediary metabolism (Coyle et al 1997, Mamus et al 2006) and supply of energy for uninterrupted carrying out of their work (McArdle et al 1999).

However, studies related to metabolic effects of pulling cycle rickshaw are rare. The present study was undertaken to assess the nutritional status of the cycle rickshaw pullers and to evaluate the effects of repeated strenuous exercise of cycle rickshaw pulling on their metabolic status.

METHODS

Study design: The present cross sectional study was undertaken in Regional Occupational Health Centre (Eastern), Kolkata in collaboration with Dept of Biochemistry, Medical College & Hospital, Kolkata. The study was approved by the Ethics Committee of both the Institutes.

Subjects: The study was carried out with the rickshaw pullers of Kolkata and surrounding areas. A list of rickshaw pullers (n=69) was chosen and prepared from different rickshaw stands. Their work experience as rickshaw pullers varied from six to sixteen years. A list of control subjects from general population other than rickshaw pullers having similar age and socio-economic status were chosen (n=33) from different localities as control group. These subjects were clinically examined. Among them 35 RP and 21 controls were selected finally for experiment after excluding those who were suffering from any acute or chronic illness detected by clinical examination. All participants were informed

about the nature, purpose and procedure of the study and written consent was obtained.

Assessment of physiological characteristics

The height and weight of the subjects were measured by a standard and calibrated stadiometer (James Scales and Engineering Company, India). Their body surface area was calculated from the equation of (Banerjee and Sen 1955) nutritional status of the subjects was assessed from their body mass index (BMI), which was calculated from body weight and height. Subjects were classified on the basis of their BMI (WHO Expert Consultation 2004), Weisell 2002, WHO 1995). Body composition of the subjects in terms of body fat percentage and lean body mass were assessed (Durnin and Rahaman 1967, Siri 1956) from their body weight and skin fold thickness at different sites of body (biceps, triceps, subscapula and supriliac) using a skinfold caliper (Holtain, UK). The maximal heart rate (HRmax) of the subjects was derived from their respective age by using the equation of (Londeree and Moeschberger 1982).

Exercise protocol of rickshaw pullers and control subjects

In order to assess the metabolic effect after strenuous exercise, each rickshaw puller was asked to pull the rickshaw six times in two phases (pre-lunch and post-lunch). Each phase consisted of three continuous cycles of 15 min work followed by rest for 15 min. They were instructed to pull a cycle rickshaw with two passengers. No restriction was imposed on the speed of the vehicle and thus they were allowed to maintain the pedalling frequency according to their own habit. The same route was used in order to avoid the variation in the road condition. All the experiments were generally started at around 10:00 hours. The range of climatic conditions during the experiments was dry bulb: 25-32°C, wet bulb:

16.5-27°C and Wet Bulb Globe Temperature index: 19.2-29°C.

Control subjects were asked to pedal a cycle ergometer (similar to RP in terms of muscle groups involved) in six cycles of 15 min work and 15 min rest. The load was adjusted to produce heart rate in the range of 70-80% of their HRmax as rickshaw pullers generally work with the same relative cardiac strain (Pradhan et al 2008).

Preparation of biological fluids for biochemical assays

Blood samples were obtained by means of sterilized disposable plastic syringes (total volume 10 ml) from antecubital veins of the study subjects maintaining proper aseptic conditions, twice according to the protocol prior to the breakfast and again after second phase of exercise. Collected blood samples were divided in four parts and aliquoted in four different vials, namely fluoride-oxalate vial for plasma glucose estimation, in EDTA vial for haemoglobin, in clot vial without any anticoagulant for serum total protein, albumin, triglyceride, cholesterol, free fatty acid, creatine kinase, creatine kinase-MB, serum urea, gamma glutamyl transferase, sodium, potassium and in 5% metaphosphoric acid for pyruvate and lactate estimation. Blood samples were kept for 30-45 minutes and centrifuged at 2000g for 15 minutes to obtain serum, plasma or clear supernatant. Urine samples were collected for 24 hours period prior to the exercise.

Biochemical assays

All biochemical measurements were done within 4-6 hours after blood sampling. Plasma glucose and serum urea were estimated by Glucose oxidase-peroxidase (GOD-POD) end point method (Trinder 1969) and enzymatic using serum urease, end point method (Taylor and Vadgama 1992) on fully automated clinical chemistry analyzer (Transasia XL-600)

by utilizing standard commercial reagents. Serum total protein and albumin quantified by biuret end point method (Silverman and Christenson 1993) and BCG dye binding end point method (Silverman and Christenson 1993) on autoanalyser (Transasia XL-600) by utilizing standard commercial Kits. Serum triglyceride, cholesterol were measured on the same instrument by Glycerol-3-phosphate oxidase-peroxidase (GPO-PAP) (Nader and Russel 2006) and Cholesterol oxidase-peroxidase (CHOD-PAP) method (Nader and Russel 2006) respectively.

Blood lactate was measured by end point method (Schumann et al. 2002) utilising lactate oxidase-paroxidase as reagent on semiautoanalyser (Erba Chem 5 V2) and pyruvate by kinetic method (Schumann et al. 2002) utilising lactate dehydronase as a reagent on the same instrument. Serum free fatty acid was analysed by FFA Quantification kit (Biovision Research Products). Fatty acids were converted to their CoA derivatives which were oxidized with concomitant generation of colour and measured photometrically (Nader and Russel 2006). Creatine kinase (CK) and CK-MB, an isoenzyme of CK, were analysed by UV kinetic (IFCC) and immunoinhibition (IFCC), UV kinetic method (Schumann et al. 2002) respectively. Serum GGT was measured by kinetic method (modified Szasz method) (Schumann et al. 2002). 24 hour urinary creatinine was measured by Jaffe's method (fixed time) (Spencer 1986).

Serum sodium, potassium and 24 hour urinary sodium, potassium were estimated by Ion Selective Electrode (ISE) (Mass et al. 1985) on Electrolyte Analyser (Medica Easylyte) according to the instruction. Blood haemoglobin was measured colorimetrically (Systronics Clinicol-631 colorimeter) by cyanomethaemoglobin method on whole blood (Van Kampen and Zijistra 1961).

STATISTICAL ANALYSIS

The data obtained were statistically analyzed using SPSS software version 17 for Windows [SPSS Inc., Chicago, USA].

RESULTS

Anthropometric and cardiovascular data and habits

Physical characteristics of the RP and control subjects are presented in Table 1. Results showed that body height, weight and body surface area were significantly higher in control subjects. BMI, Fat% and Fat (kg) values of RP and control subjects were not significantly different. However, lean body mass was significantly lower ($p < 0.01$) in RP. HR_{max} (beats/min) values of RP were similar to those of control subjects.

Both RP and controls were found to be non-vegetarian. All the subjects in this study used to take fish, eggs, chicken along with cereals and vegetables on frequent basis. Most of the RPs were smokers (88.6%) and 77% consumed alcohol. Among controls, 85.7% were smoker and 81% were addicted to alcohol.

Biochemical parameters in blood and urine

The results of biochemical analysis have been

presented in Table 2. Before exercise, there was no significant difference between RPs and controls in relation to parameters like plasma glucose, serum urea, triglyceride, cholesterol, creatine kinase (CK) and lactate pyruvate ratio. After exercise there was a significant increase in these parameters namely plasma glucose ($p < 0.001$ & $p < 0.05$), serum urea ($p < 0.001$), triglyceride ($p < 0.001$), creatine kinase ($p < 0.05$), lactate ($p < 0.001$) and pyruvate ($p < 0.001$) concentrations except cholesterol both in RPs and controls. No significant difference in CK-MB was observed in RP as compared to controls before the exercise. But after the exercise CK-MB showed significant increase ($p < 0.05$) only in control group. Gamma glutamyl transpeptidase (GGT) level were significantly lower ($p < 0.05$) in RP compared to control subjects in pre-exercise condition. There was no significant change between pre and post exercise values of both the groups.

Prior to the exercise there was no significant difference of both pyruvate and lactate in RP and control population. However, pyruvate level was significantly higher ($p < 0.001$) in control subjects when post-exercise values

Table 1. Physical characteristics of cycle rickshaw pullers and control subjects

| Parameters | Rickshaw pullers (n = 35) | Control (n = 21) | Significance Level (p <) |
|-------------------------------|------------------------------|---------------------|-----------------------------|
| Age (yrs) | 35.4 ± 5.6 | 33.8 ± 3.9 | NS |
| Body height (cms) | 161.2 ± 5.6 | 165.7 ± 3.9 | 0.01 |
| Weight (kg) | 51.4 ± 6.1 | 57.8 ± 8.6 | 0.01 |
| BSA (m ²) | 1.59 ± 0.10 | 1.69 ± 0.12 | 0.01 |
| BMI (kg/m ²) | 19.8 ± 2.5 | 21.0 ± 2.8 | NS |
| Fat% | 13.6 ± 4.3 | 14.7 ± 6.3 | NS |
| Fat (kg) | 7.1 ± 2.6 | 8.8 ± 4.7 | NS |
| Lean body mass (kg) | 44.3 ± 5.0 | 48.9 ± 5.8 | 0.01 |
| HR _{max} (beats/min) | 181 ± 3.8 | 182 ± 2.9 | NS |

Values are Mean ± SD

Table 2. Level of different biochemical parameters of rickshaw pullers and control subjects before and after exercise

| Parameters | Pre-Exercise | | Post-Exercise | |
|-----------------------|---------------------------|---------------------------|----------------------------|----------------------------|
| | RP (n=35) | Control (n = 21) | RP (n=35) | Control (n = 21) |
| Glucose (mg/dl) | 88.66±21.59 | 85.52±6.47 | 102.09±21.70 ^{a3} | 94.86±16.73 ^{a1} |
| Serum urea (mg/dl) | 25.00±6.05 | 24.81±3.06 | 30.60±7.20 ^{a3} | 29.62±3.49 ^{a3} |
| Total Protein (gm/dl) | 7.34±0.61 | 7.37±0.51 | 7.26±0.63 | 7.23±0.66 |
| Albumin (gm/dl) | 4.45±0.24 | 4.70±0.33 ^{b2} | 4.41±0.25 | 4.60±0.22 ^{c2} |
| Triglyceride (mg/dl) | 111.43±42.03 | 117.57±49.32 | 163.29±70.77 ^{a3} | 192.76±80.89 ^{a3} |
| Cholesterol (mg/dl) | 164.26±33.99 | 153.24±45.54 | 163.17±32.26 | 148.38±33.87 |
| CK (IU/lt) | 136.66±56.92 | 147.01±65.43 | 146.35±56.68 ^{a1} | 160.76±74.20 ^{a1} |
| CK-MB (IU/lt) | 11.68±6.55 | 8.80±4.40 | 12.31±6.25 | 9.95±5.14 ^{a1} |
| GGT (IU/l) | 21.41±11.07 | 29.54±18.28 ^{b1} | 21.83±8.70 | 29.38±18.88 ^{c1} |
| Pyruvate (mmol/l) | 0.06±0.02 | 0.07±0.01 | 0.11±0.04 ^{a3} | 0.16±0.04 ^{a3c3} |
| Lactate (mmol/l) | 0.84±0.16 | 0.87±0.11 | 0.95±0.16 ^{a3} | 1.06±0.24 ^{a3} |
| Lactate/Pyruvate | 14.53±4.28 ^{a3} | 12.82±1.70 ^{a3} | 9.71±3.21 ^{c3} | 6.85±2.33 |
| FFA (nmol/ micro lt) | 0.92±0.40 ^{a3b1} | 0.69±0.21 ^{a2} | 0.57±0.16 | 0.53±0.08 |
| Serum Na (mEq/l) | 137.45±4.54 | 136.69±4.78 | 139.68±3.99 ^{a3} | 138.38±3.31 ^{a2} |
| Serum K (mEq/l) | 4.48±0.52 ^{a3} | 4.42±0.73 ^{a2} | 4.12±0.45 | 4.14±0.54 |

Values are Mean ± SD

a1 = $P < 0.05$, a2 = $P < 0.01$, a3 = $P < 0.001$ (compared between Pre and Post exercise values of RP and Control)

b1 = $P < 0.05$, b2 = $P < 0.01$ (compared between Pre exercise values of RP and Control)

c1 = $P < 0.05$, c2 = $P < 0.01$, c3 = $P < 0.001$ (compared between Post exercise values of RP and Control)

Table 3. Urinary (24 h) Na, K, creatinine level and blood Hb percentage of rickshaw pullers and control subjects.

| Parameters | Rickshaw Pullers (n = 35) | Control (n = 21) | Significance Level (p<) |
|---------------------|------------------------------|---------------------|----------------------------|
| Na (mEq/l) | 156.19 ± 77.61 | 212.96 ± 73.08 | 0.01 |
| K (mEq/l) | 35.01 ± 19.48 | 42.43 ± 18.54 | NS |
| Creatinine (gm/day) | 0.92 ± 0.43 | 1.53 ± 0.78 | 0.001 |
| Hb % (gm/dl) | 14.38 ± 3.49 | 14.12 ± 1.52 | NS |

Values are Mean ± SD

were compared with rickshaw pullers. No significant difference was observed in post-exercise values of lactate concentration between the two groups, but the values were less in RPs compared to those of control group. Lactate pyruvate ratio significantly decreased after exercise in both RP and control subjects. Moreover, post-exercise value of RP was significantly higher ($p < 0.001$) than that of control values.

Free fatty acid level before exercise showed significantly higher ($p < 0.05$) value in RP than control. After exercise, the values decreased both in RP ($p < 0.001$) and control ($p < 0.01$). There was significant rise ($p < 0.05$) in serum sodium after exercise in both RPs and controls when compared with the corresponding pre exercise values. Serum potassium decreased after exercise significantly both in RPs and controls; however, there was no significant difference between these two groups after exercise.

The pre-exercise values of 24-h urinary excretion of sodium, potassium, creatinine and blood haemoglobin (%) have been presented in Table 3. Significantly low values of Na ($p < 0.01$) and Creatinine ($p < 0.001$) were observed in RPs than that of controls. There was no significant difference in haemoglobin (%) and 24 hr urinary excretion of potassium in pullers with that of the control group.

DISCUSSION

Body mass index values indicated that both RP and control groups were within 'normal' category (WHO Expert Consultation 2004). In the present study, BMI values of RP and control subjects were not significantly different.

The values of parameters like plasma glucose, serum urea, triglyceride, cholesterol, creatine kinase (CK) and lactate/pyruvate ratio indicated that all the individuals whether

rickshaw puller or controls did not suffer from any disorder of carbohydrate or lipid metabolism. Plasma glucose and serum triglycerides increased significantly ($p < 0.05$) after exercise in RPs as well as in controls. These observations are supported by earlier studies (Schenk and Horowitz 2007, Coggan 1991).

Increased post-exercise values of serum urea level indicated that there was an increased amino acid metabolism in the liver. Reduced renal blood flow causes a slight increase in the serum creatinine concentration. Although serum creatinine returns rapidly to normal level on the cessation of exercise, the increased serum urea nitrogen persists for some time (Young and Bermes 2006).

After exercise CK increased both in RP and controls whereas CK-MB showed significant increase only in control group, which may be due to adaptation of the RP to work with high intensity exercise. Reduction of cellular ATP because of high intensity exercise give rise to slight increase in the activities of non-functional serum enzymes originating from skeletal muscles such as lactate dehydrogenase, creatine kinase (Thomson et al 1975).

Increased CK-MB levels in RPs, though not statistically significant, indicate damage to skeletal muscle. The observation may be attributed to the phenomenon of 'fetal reversion'. Before exercise, the concentration of CK-MB is greater in the RPs in this study than untrained individuals (Young 1979, Apple and Jaffe 2006). The mechanism responsible for increased CK-MB is thought to be caused by the regeneration process of muscle, with re-expression of CK-MB genes similar to those found in the heart thus giving rise to increased CK-MB levels in skeletal muscles (Apple and Billadello 1994). Gamma glutamyl transpeptidase (GGT) level were significantly

higher in control group than that of RP both before and after exercise, but the values obtained were within reference interval (Panteghini et al 2006).

Although there was increase in lactate concentration in RPs as well as in control subjects after exercise, it was far less than that of control population indicating that rate of gluconeogenesis is more in RP population than that of control. The result of the present study is supported by earlier observations (Sacks 2006) where it is reported that strenuous exercise increases lactate concentration significantly from an average concentration. The lactate response to exercise is reduced in trained athletes compared with untrained individuals (Young and Bermes 2006)

Free Fatty Acid level was significantly higher ($p < 0.05$) in RP than that of controls before exercise. Fatty acid mobilizations as well as fatty acid oxidation by muscle were increased in the RPs as adaptive response to the strenuous exercise associated with their job. After exercise, free fatty acid concentration significantly decreased ($p < 0.05$) in both the groups, suggesting increased glycolysis than that of free fatty acid oxidation, because all the subjects took meal before performing exercise. Increased intracellular glucose concentration facilitates esterification of fatty acid to neutral fat (i.e., triglycerides). Therefore, the breakdown of stored fat to fatty acid decreases, leading to decrease in circulating fatty acid concentration (Coyle et al 1997, Mamus et al 2006)

Increased serum sodium after exercise in both RPs and controls when compared with the corresponding pre exercise values may be due to renal or extra-renal loss of hypo-osmotic fluid and Na^+ re-absorption at the distal tubule (Sacks 2006). Serum potassium level decreased after exercise both in RPs and

controls; this may be due to consequences of glucose transport in the cells. Shifting of K^+ from extracellular fluid into the cells as H^+ moves in the opposite direction can also be possible cause for decreased K^+ after exercise (Klutts and Scott 2006)

CONCLUSIONS

In rickshaw pullers, increase in lactate was far less than that of controls suggesting higher exercise tolerance. Lactate/pyruvate ratio less than 25 both in pre and post exercise state of both groups suggested normal gluconeogenesis. Low values of 24-h urinary Na and creatinine prior to exercise indicated that renal reabsorption capacity in RP were higher perhaps resulting from adaptive response to altered metabolic status.

ACKNOWLEDGEMENTS

The investigators sincerely express their gratitude to Department of Science and Technology, Govt. of India for their sponsorship and financial support to carry out this project. The authors are thankful to the Director, National Institute of Occupational Health (NIOH), Ahmedabad and the Officer in Charge, Regional Occupational Health Centre (E), Kolkata for their guidance and support to carry out the study. The investigators thank the cycle rickshaw pullers and other subjects for their participation and cooperation to conduct the study.

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