

Oxygen uptake of the freshwater crab *Paratelphusa spinigera*, Wood Manson (1871), at different seasons of the year

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Abstract

The rate of respiration is a useful and sensitive measure of an organisms' daily expenditure of energy. Oxygen consumption rate of freshwater crab *Paratelphusa spinigera* was studied from December 2017 to August 2018. The experiments were conducted by the cylindrical glass respirometer over winter ($16 \pm 1^\circ\text{C}$) and summer ($34 \pm 1^\circ\text{C}$) seasons with the aim of estimating oxygen consumption. Data were analyzed from logarithmic transformations of body weight (W) and oxygen uptake (VO_2) expressed by the allometric equation $\text{VO}_2 = aW^b$. The total oxygen uptake increased with 0.070 power of body weight in winter and with 0.071 power of body weight in summer. The specific oxygen consumption decreased with 0.929 power of body weight in winter and with 0.927 power of body weight in summer. The correlation coefficient (r) between oxygen uptake and body weight was positive significance i.e. 0.99599 ($p < 0.001$) in winter and 0.9879 ($p < 0.01$) in summer. The study was concluded that the oxygen consumption rate was increased with increasing body mass of the crab. But the rate of metabolism (the specific oxygen uptake rate) was decreased with increasing mass at both summer and winter. The specific metabolic rate is higher in smaller than in larger crabs.

Keywords: temperature, respirometer, correlation, dissolved oxygen

1. Introduction

Fresh water crabs (*Paratelphusa spinigera*), bimodal breathing crustaceans, represent a stage in the transition from the aquatic to terrestrial habitat. They have both gills and enlarged & vascularised branchiostegites (branchial chamber) which are used for respiration in ambient habitats (water and land). They can intake dissolved oxygen in water through well vascularised gills. The received oxygen is utilized in cellular respiration to release energy on which their life processes depend. Thus growth and production of the crabs in aquaculture depend on amount of oxygen taken by them. A detailed study was made on radical adaptation of all Brachyura with respect to mode of respiration (Lutz, 1969) [24]. Taylor and Greenaway (1979) [34] worked on gill structure of terrestrial crab *Holthuisana* spp. Copeland (1963) [9] discovered possible osmoregulatory cells in crab's gills. The gill chambers of the coconut crab (*Birgus labto*) generally held enough water to guarantee high humidity of the chambers (Edney, 1960; Mill, 1972 and Taylor *et al.*, 1973) [12, 26, 33]. The metabolic rate on relation to size of body of many poikilotherms has been studied by several investigators. Zeuthen (1953) [41] and Weymouh *et al.* (1944) showed that weight specific oxygen uptake decreased with increasing body size. Individual rate of oxygen consumption of the freshwater crab (*Elamenopsis kempfi*) increased with increasing body mass at all temperatures for males, females and ovigerous females (Ali *et al.*, 2000) [3]. Alekseeva and Zotin (2001) [2] worked about the influence of temperature and other environmental factors on the rate of oxygen consumption. Islam *et al.* (2003) [16] made a detailed study on bimodal oxygen uptake and gill structure of *Paratelphusa spinigera*. The oxygen consumption rate was positively correlated with the body weight of the crabs but was inversely related to its unit

weight per hour (Inyang and Nicholas, 2004). Katsanevakis *et al.* (2007) [20] worked on oxygen consumption of the semi-terrestrial crab (*Pachygrapsus marmoratus*) in relation to body mass and temperature. Bett and Vinatea (2009) [7] reported temperature, salinity, shrimp size and the interaction of these parameters significantly affect the specific oxygen consumption. The rate of respiration of *Balanus glandula* was increased exponentially with body mass to the 0.66 power (Gilman *et al.*, 2013) [13]. Bansode (2015) [4] made a detailed study on daily rhythm of oxygen consumption in fresh water crab (*Barytelphusa jaquemontii*).

Measurement of rate of respiration of aquatic animals is essential for aquaculture. Oxygen uptake by the crab *Paratelphusa spinigera* was not done in Biratnagar. So the information obtained from this study will help to know some ideas about stocking density of the crab in water bodies of Biratnagar, eastern part of Nepal.

2. Materials and Methods

Study area

The study was carried out in Biratnagar (Alt. 72 MSL, $26^\circ 28' 60''$ N & $87^\circ 16' 60''$ E) located in Morang district of Province 1, Nepal. It is the busiest and 2nd largest industrial city with population of 2, 40,000 & area of 103.88 km².

Specimen collection

Live specimens of the crab *Paratelphusa spinigera* of different body weight were captured by hands from rice fields, ditches, ponds and canals of Biratnagar with the help of local farmers in winter season (December, January and February) and summer (June, July and August) season from December, 2017 to August, 2018.

The collected specimens were acclimatized in a large glass

aquarium inside Zoology laboratory of Post Graduate Campus, Biratnagar for a week. The temperature acclimation was similar to that of the natural environment at each sampling date. Seven specimens were taken for experiment in each season. The experimented crab was starvated for 12 hours before experimentation. The crabs were weighed before introduced into the experiment. Atmospheric temperature, room air temperature, and temperature and pH of water were also recorded.

Measurement of oxygen consumption

A flow-through cylindrical glass respirometer (24 cm long and 7 cm in diameter) of 722 ml volume, was used in order to measure the inflow and the out-flow water. The oxygen content of the inspired and expired water was measured by means of the Winkler’s method (Welsh, 1948). The constant flow of water maintained in experiment per hour was 600 ml. Flow of water was controlled by using metal clops. The oxygen uptake per unit time and body weight was determined by the difference of dissolved oxygen of inspired and expired water and body weight of the crabs. A large water tank with constant water level was connected to the respirometer. The tank was connected to chlorine free tap water. At first rate of flow of water was high and then slowed down. The crab was acclimatized in the respirometer for one hour before experimentation. Visual disturbance was avoided by a black cloth cover over the respirometer. Both ends of the respirometer were connected to the conical flasks from where inspired and expired water were collected for measurement of concentration of the dissolved oxygen content using Winkler’s volumetric method (Welch, 1948). Oxygen consumption per unit time (mlO₂/hr) and per unit body weight (mlO₂/g/hr) was calculated by the differences of dissolved oxygen levels between inspired and expired water and body weight of the crab. At every half an hour interval, three readings were taken and then an average value was calculated out of the three readings.

Regression analysis after logarithmic transformation was done to show regression coefficient between oxygen uptake and body weight and also between specific oxygen consumption and body weight. The correlation coefficient and estimated value were calculated by power equation. The expression of the relationship between oxygen uptake and body weight was made by the following equation:

$$VO_2 = aW^b$$

or
 $\text{Log } VO_2 = \text{log } a + b \text{ log } W.$

Where, VO₂ = oxygen uptake per unit time a = intercept
 b = regression coefficient W= weight of crab

Statistical analysis

Coefficient of correlation between body weight and oxygen uptake per unit time with number of crabs collected were performed by using Karl Pearson’s method. The coefficient of correlation was calculated by using excel software.

3. Results

Winter season

The rate of oxygen uptake, temperature and pH is presented in Table 1. During winter the atmospheric temperature ranged from 15 to 19° C whereas the water temperature and ambient temperature were ranged from 16 to 20° C and 16 to 19° C, respectively. The pH of water was 7. The weight of crab ranged from 5g to 24g.

The oxygen content of ambient water (DO) ranged from 4.83 to 6.04 mg/l.

Table 1: Data of oxygen uptake in relation to body weight in winter

Body wt. (g)	Atm. Temp. (°C)	Ambient temp. (°C)	Oxygen uptake		
			mlO ₂ /hr	mlO ₂ /g/hr	mlO ₂ /kg/hr
5.00	15	16	1.21	0.24200	242.00
7.00	16	17	1.23	0.17571	175.71
11.00	17	17	1.27	0.11545	115.45
16.00	17	18	1.30	0.08125	81.25
20.50	18	18	1.33	0.06488	64.88
22.40	18	19	1.34	0.05982	59.82
24.00	19	19.5	1.35	0.05625	56.25
Avg=15.13	Avg=17.14	Avg=17.72	Avg=1.290	Avg=0.1136	Avg=113.6240

Body weight of crabs(X), oxygen uptake(Y), logX and logY are presented as the winter parabolic chart for statistical analysis (Table 2). Body weight of crabs ranged from 5g to 24g and oxygen uptake (mlO₂/hr) ranged from 1.21 to 1.35. Value of logX, logY and their square and products are increased as increasing body weight.

Table 2: The Winter Parabolic Chart.

S. N	X	logX	logX ²	Y	Logy	logY ²	logX.logY
1	5.00	0.69897	0.488559	1.2	0.08278	0.00685335	0.0578642
2	7.00	0.84509	0.714190	1.2	0.08990	0.00808290	0.0759785
3	11.00	1.04139	1.084499	1.2	0.10380	0.01077527	0.1081007
4	16.00	1.20412	1.449905	1.3	0.11394	0.01298232	0.1371974
5	20.50	1.31175	1.720698	1.3	0.12385	0.01533931	0.1624633
6	22.40	1.35024	1.823169	1.3	0.12710	0.01615568	0.1716232
7	24.00	1.38021	1.904982	1.3	0.13033	0.01698695	0.1798884
Sum (Σ)	105.90	7.83179	9.186004	9.0	0.77172	0.08717581	0.8931160

The relationship between oxygen uptake (mlO₂/hr) and body weight (g) at temperature 16 ± 1° C

Oxygen uptake in the crabs ranged from 1.21 to 1.35 (mlO₂/hr) within the weight range of 5g to 24g (Table 1). The slope of regression line from logarithmic equation was 0.070. The oxygen uptake (mlO₂/hr) increased from 1.21 to 1.35 with increasing body weight from 5g to 24g by a power of 0.070. (Fig.1). There was positive significance between oxygen consumption and body weight with the correlation coefficient (r) equal to 0.99599 (p<0.01). The increased oxygen consumption exhibited an exponential relationship with increasing body. The value of regression coefficient and intercept was 0.070 and 1.076 respectively.

Here, VO₂ = aW^b

Or, Log. VO₂ = log.a + b log.W = log 1.076 + 0.070logW = 0.031812 + 0.070logW

Or

$$VO_2 = 1.076W^{0.070}$$

The estimated value for 1 g, 10 g, 10 g and 1000 g crab was 1.076, 1.26418, 1.48529 and 1.74506 respectively.

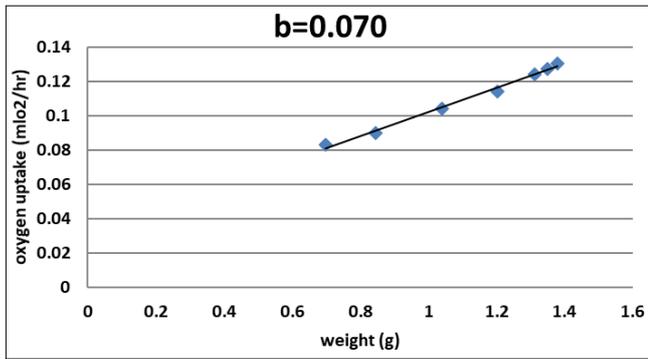


Fig 1: Log-log plot of the relationship between oxygen uptake (mlO₂/hr) and body weight (g) at 16 ± 1°C in *P. spinigera*.

Relationship between body weight and oxygen uptake per unit body weight (mlO₂/g/hr) at 16 ± 1°C

The weight specific oxygen uptake (mlO₂/g/hr) decreased from 0.24200 to 0.05625 with increasing body weight from 5g to 24g (Table 1). The slope of regression coefficient by log-log plot of the relationship between specific body weight and oxygen uptake was negative i.e. -0.929. Weight specific oxygen uptake (mlO₂/g/hr) was decreased with increasing body weight by power of 0.929 (Fig. 2). The correlation coefficient obtained by power equation was negative. It is equal to -1 (p<0.01) which showed significance.

Here,

$$VO_2 = aW^b = 1.076W^{-0.93}$$

The estimated value for 1g, 10g, 100g and 1000g crab were 1.076, 0.12641, 0.01485 and 0.00163 respectively.

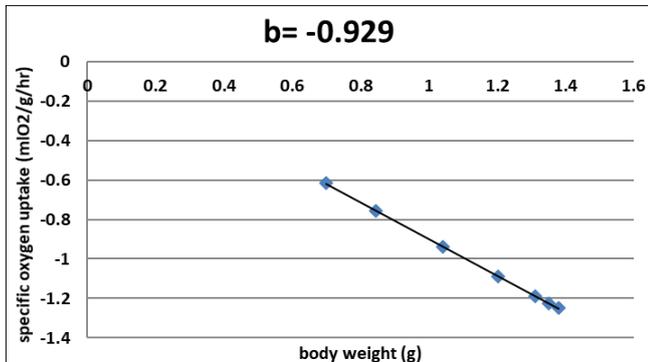


Fig 2: Log-log plot of the relationship between body specific oxygen consumption (mlO₂/g/hr) and body weight (g) at 16 ± 1°C in *P. spinigera*.

The oxygen uptake with intercept, slope, and correlation coefficient in *P. spinigera* during winter was shown in Table 3.

Table 3: Oxygen uptake showing intercept, slope and correlation coefficient (r) in *P. spinigera* during winter.

Body wt. Vs O ₂ consumption	Intercept (a)	Slope (b)	Correlation Coefficient (r)
VO ₂ (mlO ₂ /hr)	1.076	0.070	0.99599
VO ₂ (mlO ₂ /g/hr)	1.076	-0.929	-1

Summer season

The rate of oxygen uptake and temperature were presented in Table 4. During summer the atmospheric temperature

ranged from 30 to 35° C whereas the ambient temperature was ranged from 30 to 34° C. The pH of water was 7 and water temperature was ranged from 30 to 34° C. The weight of crabs ranged from 10g to 32g. The oxygen content of ambient water (DO) ranged from 4.23 to 5.24 mg/l.

Table 4: oxygen uptake in relation to body weight in summer

Body wt.(g)	Atm. Temp. (°C)	Ambient Temp. (°C)	Oxygen uptake		
			ml O ₂ /hr	ml O ₂ /g/hr	ml O ₂ /kg/hr
10	30	31	1.2900	0.1290	129.0000
12	31	30	1.3200	0.1100	110.0000
16	32	33	1.3300	0.0831	83.1250
20	33	33	1.3500	0.0675	67.5000
26	33	34	1.3800	0.0531	53.0769
30	34	33	1.4000	0.0467	46.6667
32	35	34	1.4100	0.0441	44.0625
Avg.=20.86	Avg.=32.57	Avg.=32.57	Avg.=1.3543	Avg.=0.0762	Avg.=76.2044

Body weight of crabs(X), oxygen uptake(Y), logX and logy are presented as the summer parabolic chart for statistical analysis (Table 5). Body weight of crabs ranged from 10g to 32g and oxygen uptake (mlO₂/hr) ranged from 1.29 to 1.41. Value of logX, logy and their square and products are increased as increasing body weight.

Table 5: Summer parabolic chart.

S.N.	X	logX	logX ²	Y	Logy	logy ²	logX. Logy
1	10	1	1	1.2900	0.1106	0.01223	0.11059
2	12	1.079181	1.164632	1.3200	0.1206	0.014538	0.130121
3	16	1.20412	1.449905	1.3300	0.1239	0.015339	0.149132
4	20	1.30103	1.692679	1.3500	0.1303	0.016987	0.169568
5	26	1.414973	2.00215	1.3800	0.1399	0.019566	0.197925
6	30	1.477121	2.181887	1.4000	0.1461	0.021353	0.215849
7	32	1.50515	2.265476	1.4100	0.1492	0.022266	0.224597
Sum(Σ)	146	8.981575	11.756729	9.4800	0.9205752	0.122280	1.197782

Relationship between body weight (g) and oxygen uptake per unit time (mlO₂/hr) at temperature 34 ± 1°C

The relationship between body weight (g) and oxygen uptake (mlO₂/hr) at temperature 34 ± 1°C is shown in Fig. 3.

As winter season, the summer has also the oxygen uptake increased with increasing body weight of the crabs. The increased oxygen consumption exhibited an exponential relationship with increasing body weight. A straight line was obtained when the log-log plot of the oxygen uptake in relation to body weight were filled by linear equation method (Fig.3). The slope of regression line was 0.071. The oxygen uptake (mlO₂/hr) increased from 1.29 to 1.41 with increasing body weight from 10g to 32g by a power of 0.071. The correlation coefficient 'r' between oxygen uptake per unit time (mlO₂/hr) and body weight was 0.9879 (p<0.01). The relationship between oxygen uptake per unit time (mlO₂/hr) and body weight was represented by regression equation in the form of VO₂ = aW^b = 1.096W^{0.071} Or

$$\text{Log VO}_2 = 0.039811 + 0.071 \text{LogW}$$

The estimated value for 1g, 10g, 100g and 1000g were 1.096, 1.290656, 1.519884 and 1.789824 respectively.

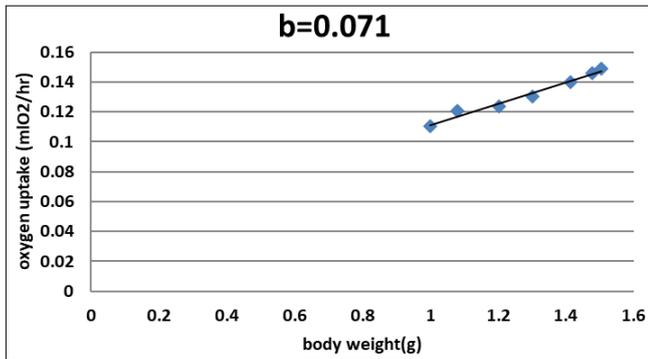


Fig 3: Log/log plots showing the relationship between oxygen uptake and body weight (mlO₂/hr) at 34 ± 1°C in *P. spinigera*.

Relationship between body weight and rate of oxygen uptake per unit body weight (mlO₂/g/hr) at 34 ± 1°C

The weight specific oxygen uptake (mlO₂/g/hr) decreased from 0.1290 to 0.0441 with increasing body weight from 10g to 32g (Table 4). A straight line was obtained when the data for weight specific oxygen uptake rate were plotted against body weight on log-log co-ordinate (Fig.4). The slope of regression coefficient was negative i.e. -0.927. Weight specific oxygen uptake (mlO₂/g/hr) was decreased with increasing body weight by power of 0.927 (Fig. 4). The correlation coefficient obtained by power equation was negative. It is equal to 0.9994 (p<0.01) which showed negative significance.

Here,

$$VO_2 = aW^b = 1.094W^{-0.92}$$

Estimated value of 1g, 10g, 100g and 1000g of crabs were 1.094, 0.1315, 0.0158 and 0.0012 respectively.

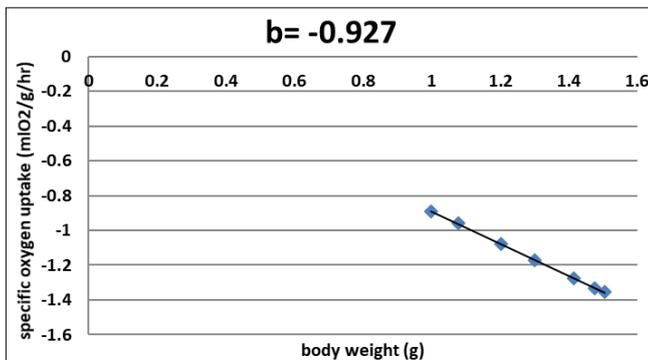


Fig 4: Log/log plots showing the relationship between weight specific oxygen consumption (mlO₂/g/hr) and body weight (g) at 34 ± 1°C in *P. spinigera*.

Oxygen uptake, intercept, slope and correlation coefficient in the crab during summer was summarized in Table 6.

Table 6: Oxygen uptake, intercept, slope and correlation coefficient in summer.

Body wt.(g) Vs oxygen consumption	Intercept(a)	Slope (b)	Correlation coefficient(r)
VO ₂ (mlO ₂ /hr)	1.096	0.071	0.9879
VO ₂ (mlO ₂ /g/hr)	1.094	-0.927	-0.9994

In conclusion, the outcome of the study on the oxygen uptake and metabolic weight exhibits that the rate of oxygen uptake (mlO₂/hr) in the crab varies on size of animals. The oxygen uptake per unit time is more in larger animals and

less in smaller ones both in winter and summer seasons. But metabolic rate (mlO₂/g/hr) is comparatively less in larger specimens in both seasons. The total oxygen uptake increased with 0.070 power of body weight in winter and with 0.071 power of body weight in summer. The metabolic rate of oxygen consumption was decreased exponentially with body weight to 0.929 power in winter and to 0.927 power in summer.

4. Discussion

Ambient temperature in winter and summer were 16 ± 1°C and 34 ± 1°C respectively. The pH in both season remained same i.e. 7. The dissolved oxygen in winter was greater (4.83 to 6.04mg/l) than summer (4.23 to 5.24 mg/l). It is agreed to Wetzel (2001) [37] “Cold water can hold much dissolve oxygen as warm water.”

The present study showed that the oxygen consumption rate increased with increasing body mass of the crab. It is in accordance to the result of Diawol *et al.* (2016) [11], who stated that oxygen uptake of freshwater decapods varied according to the animal’s weight. The rate of oxygen consumption is generally increased with increasing body mass of the crab at all temperature (Ali *et al.*, 2000) [3] is also similar to this study. In the present study, the rate of metabolism (the specific oxygen uptake rate) was decreased with increasing mass at both summer and winter. This result is similar with earlier studies on aquatic invertebrates (Kleiber, 1947; Zeuthen, 1947, 1953, 1970; Winberg, 1956; Marsden, 1979; Al-Dabbagh and Marina, 1986) [22, 42, 39, 25, 1]. The study presumed the rate of respiration is increased in all group of *P. spinigera* with increase of temperature which is compatible with those acquired for other invertebrates (Kinne, 1970; Ivleva, 1980; Ali *et al.*, 2000) [21, 17, 3].

The oxygen uptake depends upon various abiotic and biotic factors (Imabayashi and Takahashi, 1987) [14]. Of these factors, body weight is one which plays a very important role. The slopes of the regressions of the rate of oxygen consumption and wet body weight of the crab (*P. spinigera*) from winter to summer, ranged from 0.070 to 0.071. This result differed from other workers. The slope of the regressions of the rate of oxygen consumption and dry body weight of aquatic crustaceans were 0.81 (Winberg, 1950; 1956); 0.75 (Sushchenya, 1970) [32], 0.66 (Gilman *et al.*, 2013) [40, 13] and 0.5 to 1 (Klein-Breteler, 1975; Marsden, 1979) [23, 25]. These variations are due to several factors like body weight, gonad size, activity and nutritional status; which are either not considered or their effects are minimized to various degrees and are inevitable in the laboratory work. Environmental time i.e. day or month and body size affect the degree of voluntary activities which ultimately influence the internal metabolic rate. Smaller animals have higher rate of metabolism than larger animals (Bansode, 2015) [4]. This finding was agreed to the finding of the present study. These are fully noticeable through the scattering of the point of the respiratory relationships. Rao and Bullock (1954) [29] highlighted that the meticulous evaluate of the slope depends largely on minimizing of the scattering of these points to the nearest possible limits. In fact, if this statement attained, would provide a great value in the physiological studies.

The rate of oxygen consumption in any animals is connected to the vastness of the energy requirements of the animals. It depends on animal’s body weight (Bertalanffy, 1957) [6] and environmental factors (Dejours, 1976) [10].

The value of exponent relating to oxygen uptake per unit time in *P. spinigera* was never greater than 1. It means the weight specific oxygen uptake ($\text{mlO}_2/\text{g/hr}$) decreased with an increase in body weight. This result agreed to Fry (1957) reported the high rate of oxygen uptake per unit body weight in small fish has their high metabolic rate. Decreased weight specific oxygen uptake may be due to sluggishness of larger animal than smaller ones (Roy, 1983). Another possible cause may be due to enlarge in inert tissue like skeleton, blood water diffusion barrier and metabolites in larger animals. Several workers have suggested different exponent values to state the relationship between oxygen uptake and body weight (Brody, 1945; Job, 1955; Parvatheswara Rao, 1960; Beamish and Mukherjee, 1964 and Kamler, 1976)^[8, 18, 29, 5, 19]. The exponent values of more than 1 have also been reported by several workers (Ware and Igram, 1979; Munshi *et al.* 1978)^[35, 27]. An exponent greater than 1 means that they predict the weight specific oxygen uptake increases with increasing body weight.

The intercept value for oxygen uptake in *P. spinigera* in winter was 1.0760 and in summer was 1.09624. This study concludes that small crabs are more active. They ventilate more to extract the required oxygen. The metabolic rate is more in smaller specimen than larger ones. Perhaps large crab has sluggish movement and less energy is expended in search of foods as the crab gets food and shelter from where it clings on the branches of the water plants. The temperature also found to play a great role for the change of dissolved oxygen in water and the rate of oxygen uptake extracted by the similar weight of crab (Bansode, 2015)^[4]. The rate of oxygen uptake was greater in winter than summer. It indicates the oxygen uptake might be increased as temperature increased. This result is similar with that of Siikavuopio and James (2015)^[31], who stated that oxygen consumption was significantly affected by temperature and increased with increasing temperature.

Smaller animals lean to have higher per gram basal metabolic rates than larger animals. This may be because of the higher metabolic rate of small animals needs a greater delivery of oxygen to tissues around the body as well as the smaller animals may have a greater surface area to volume ratio, so more heat is lost. Oxygen consumption represents the physiological state of metabolic activity and it may be an indicator of metabolic stress. The environmental factors may induce stress to exposed animals. The cost of transport can be minimized by transporting them in chilled air rather than vivier lorries (Robson *et al.*, 2007)^[30].

5. Conclusion

The present study stated oxygen consumption rate was increased with increasing body mass of the crab. But the rate of metabolism (the specific oxygen uptake rate) was decreased with increasing mass at both summer and winter. The study concluded that oxygen consumption of the crab depends on its size/age. Oxygen consumption rate of the crabs and dissolve oxygen in the water can predict the period for transportation. So the significance of the work is to know the how much oxygen is required in the transportation of different ages/sizes of the crabs.

6. Acknowledgment

We would like to give a special thank to Dr. Bharat Raj Subba who gave his precious suggestions during research work. We would like to thank to Hari Maya Shrestha for

helping in typing work. We would like to express our deep sense of gratitude to fisherman Jageshor who helped to collect the crabs from the study areas.

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