WATER, ENERGY AND NITROGEN UTILIZATION BY LARVAE OF FIVE SPECIES OF LEPIDOPTERA FEEDING ON *PRUNUS SEROTINA* LEAVES

by

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Submitted in Partial Fulfillment of the Requirements
for the Degree of
Master of Science
in the
Biology
Program

Lawrence Alfred Schroeder August 10, 1977
Adviser Date

Dean of the Graduate School Aug 13, 1977
Dean of the Graduate School Date

YOUNGSTOWN STATE UNIVERSITY

August, 1977

ABSTRACT

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Dry mass, energy, nitrogen and water budgets were determined for five species of lepidopteran larvae fed on freshly collected Prunus serotina leaves. Significant interspecific differences were found for all efficiencies of utilization with the exception of gross nitrogen utilization efficiency. Net efficiency of energy utilization correlated only with gross nitrogen utilization efficiency and with rate of production of metabolic water. Gross nitrogen utilization efficiency correlated positively with rate of growth and with water availability and negatively with both gross and net water utilization efficiencies. It was hypothesized that energy efficiencies are limited by genetically controlled, interspecific, adaptive differences while nitrogen efficiency is limited by environmental factors more universally applicable according to life strategy. It was also suggested that the observed data were consistent with the hypothesis of energy efficiency maximization.

ACKNOWLEDGEMENTS

I am indebted to Dr. Lauren Schroeder. He willingly helped with all phases of my work, but more important perhaps, he has the rare talent for providing encouragement and challenge in proper measure. My husband, Ralph, not only got me started, but often kept me going. My mother and daughter were likewise supportive. To all of these I now offer the thanks that I may have neglected along the way.

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INTRODUCTION

Much current debate in ecology revolves about the concept of maximization, and there is controversy over which processes may be maximized either evolutionarily or successionally. Odum and Pinkerton (1955) proposed that evolution tended toward maximization of power output, which occurs at relatively low efficiencies. Others (MacArthur, 1955; Morowitz, 1968; Margalef, 1968; 1975; Fox, 1971) have suggested that both evolution and succession would tend toward maximization of stability on the principle that more stable systems would by definition be longer lived and succeed over less stable systems. More recently, Schroeder (1977_b) used thermodynamic principles to argue for the maximization of complexity which, under these same thermodynamic principles, must be positively related to both numerical diversity and food web complexity. He further proposed that maximization of complexity could result only from maximization of individual energy utilization efficiencies.

Approaching from the opposite direction, Gates (1968) said, "If the environment is to influence an organism it must do so by energy transfer . . . All interactions can be reduced to an energy basis." Waldbauer (1968) agreed to the importance of energy in ecological studies and proposed that insects, being a major component

of almost all terrestrial and fresh water communities, might be a productive area for study. Since insect energy efficiencies are usually considerably lower than Schroeder's (1977_b) theoretical maxima (Waldbauer, 1968), it would be interesting to ascertain which environmental or internal constraints might operate as limiting factors.

One possible limitation to the efficiency of herbivorous insects might be the secondary substances produced by their food plants. Gordon (1959), Waldbauer (1968) and Whittaker and Feeny (1971) proposed that oligophagous insects, exposed to fewer toxic secondary plant substances, could devote less energy to elaboration of detoxication enzymes than polyphagous insects which would be exposed to a wider range of toxic substances. This would result in higher energy utilization efficiencies among oligophagous insects as compared with polyphagous insects. If efficiency is to be maximized, insects would tend to evolve the narrowest food niche compatible with the extant conditions of availability and certainty of their food plants. Since ecological succession tends toward more stable, longer lived plant communities, the insect herbivores of later successional stages should have narrower food niches resulting in higher utilization efficiencies. Working with Lepidoptera, Schroeder (1976; 1977_a) failed to find support for this hypothesis and concluded that, at the minimum, efficiency differences

possibly due to degree of food plant specialization were complicated by other aspects of life strategy.

Feeny (1976) hypothesized that plant species differ in the nature of their chemical defenses. He argued that "unapparent" plants (ephemeral, widely spaced species, generally of early successional stages) benefit by being different from neighboring species. They therefore tend to produce small quantities of specific defense chemicals which are highly effective in small amounts against non-adapted species, but which are subject to rapid detoxication by adapted herbivores. "Apparent" plants (long-lived, abundant species, generally woody and of later successional stages), since they are likely to have as neighbors other members of the same species, gain little from interspecific differences. These produce larger amounts of less specific defense chemicals which are effective in proportion to their concentration and are likely to inhibit feeding or growth or both. These chemicals are less susceptible to detoxication mechanisms.

Futuyma (1976) concluded that due to consideration of escape strategies of plants, the very basis of the theory of oligophagous-polyphagous efficiency differences was in error. He argued that specialists tend to feed on "unapparent" plants, while generalists feed on "apparent" plants. Since the food plants of generalists are more nearly alike in their secondary substances than are the food plants of

specialists, polyphagous insects may not be exposed to a wider range of toxic substances than are oligophagous insects.

Differential accumulation of nutrients is another possible limiting factor for utilization efficiencies. This effect may be particularly pronounced among herbivores whose tissue nitrogen concentration may be several times that of their food plants. Slansky and Feeny (1977) found that larvae of the cabbage butterfly consumed greater amounts when fed on low nitrogen plants than when fed on high nitrogen plants. Thus, the rate of nitrogen accumulation was stabilized at the expense of dry mass and energy utilization efficiencies.

Just as nitrogen must be concentrated by herbivorous insects, so must water be concentrated by those species which obtain all or most of their water from food plants. Although the depressing effect on growth and development of low atmospheric or food water has been well documented (Fraenkel and Blewett, 1944; Bursell, 1964; Wigglesworth, 1965), as has the ability of insects to produce drier feces under water stress (Edney, 1957; Bursell, 1964; Waldbauer, 1964; 1968; Schroeder, 1968), little attention has been directed toward the effect of water availability on utilization efficiencies. Using cecropia larvae under four different combinations of relative humidity and leaf water supplementation, Scriber (1977) has

shown that utilization efficiencies of dry matter, energy and nitrogen, as well as growth rate, vary directly with the percentage of water in the leaves used as food.

The work reported in this paper was an attempt to study the effect of leaf water, as it varies naturally during the season, on the efficiencies and growth of certain lepidopteran larvae. This is important because it is possible that those species which occur later in the year may have adapted so that their efficiencies remain high in spite of low leaf water.

MATERIALS AND METHODS

Collecting and Rearing Techniques

Lepidopteran larvae were collected by Dr. Lauren Schroeder and myself from black cherry trees (Prunus serotina) in Lawrence County, Pennsylvania and Mahoning County, Ohio. Larvae were placed individually into appropriately sized glass vials divided longitudinally by size 50 mesh brass screening into rearing and control chambers. Vials were closed with perforated plastic caps.

Larvae were reared in an environmentally controlled chamber with a 14 hour photoperiod at 27°C, and 75% RH.

Fresh food leaves were gathered daily from black cherry trees in Lawrence County, Pennsylvania or Mahoning County, Ohio, and transported to the laboratory in sealed plastic bags. If surface water was evident, this was removed by blotting on paper towels. Tips, bases, and midveins of excised leaves were removed. The two remaining mid-sections were assigned randomly to food or control categories and weighed, using a Sartorius semimicro balance, to a precision of 0.2 mg.

Control leaves from the previous day were removed from the vial, reweighed, labelled, placed in aluminum weighing pans and dried in a partial vacuum over $CaCl_2$ at 60°C for 48 hours. Uneaten food (residual) and fecal

pellets were removed, labelled, placed in pans, and similarly dried. Larvae were weighed and returned to the vial along with the newly weighed food and control leaves.

At the onset of pupation, when the larvae became inactive or began to spin a cocoon, they were weighed, killed and dissected to remove any unassimilated food from the gut. This was added to the pan with the fecal pellets. Larvae were placed in separate pans and labelled for drying.

Drying

All drying was done over $CaCl_2$ for 48 hours at 60°C in a partial vacuum.

Determinations of Water Content

Leaf Water

Water content of leaves fed to larvae was estimated daily for each larva from the water content of control leaves. Preliminary experiments had shown that leaf water loss was linear with time following a brief initial period of rapid loss. Mean fresh weight of the leaves was therefore taken as the arithmetic mean of the initial weight and the weight at the end of the feeding period. Fractional water content was obtained by:

$$H_2O_{(leaf)} = \frac{\text{mean fresh wt.} - \text{dry wt.}}{\text{initial wt.}} \qquad (1)$$

Water content of leaves fed to larvae was obtained by multiplying the initial weight of the leaves by the decimal fraction given in Equation (1).

Larval Water

Water content of the initial larvae was calculated as live weight multiplied by 0.85. Although water content of caterpillars varies between 80 and 95% (Scriber, 1975), the initial weights of most larvae were quite small and the error introduced here is correspondingly small. The water content of terminal larvae was determined directly as the difference between live weight and dry weight.

Fecal Water

Freshly deposited fecal pellets were collected at 10 minute intervals and weighed. Since mean elapsed time between deposition and weighing was 5 minutes, pellets were reweighed five minutes after the original weighing, and the five minute weight loss was added to the original weight as a correction factor. Pellets were dried and reweighed to determine water content, Equation (2).

$$H_2O_{(feces)} = \frac{\text{fresh wt.} - \text{dry wt.}}{\text{dry wt.}} \qquad (2)$$

This was done a minimum of four days weekly for each larva. Water content of fecal material was calculated from observed dry weight and the decimal fraction given in Equation (2).

Metabolic Water

Water produced from metabolic processes was calculated according to Equation (3).

$$M = 0.55C_c + 0.20P_c = 0.49F \quad (3)$$

where: M = metabolic water in grams

C_c = carbohydrate catabolized (grams)

P_c = protein catabolized (grams)

F = fat synthesized (grams)

The coefficients represent the weight, in grams, of water obtained per gram of carbohydrate or protein metabolized or of fat synthesized (Chefurka, 1965_a and 1965_b).

Ingested protein was calculated as 6.25 times the total nitrogen ingested ($100 \times N\%_{(leaves)} \times$ dry wt.). Larval protein was similarly calculated from larval nitrogen percentages and dry weights of growth. Catabolized protein was the difference between ingested protein and stored protein.

Fat synthesized was obtained by simultaneous solution of the dry weight and energy equations, (4) and (5), respectively.

$$W = C_l + P_l + F \quad (4)$$

where: W = weight of larva

C_l = weight of carbohydrate in larval tissue

P_l = weight of protein in larval tissue

F = weight of fat synthesized

$$Cal = 3500 \frac{cal}{g} \times C_1 = 4500 \frac{cal}{g} \times P_1 = 9300 \frac{cal}{g} \times F \quad (5)$$

where: Cal = total energy content of the larva

$3500 \frac{cal}{g}$ = energy equivalent for carbohydrate

$4500 \frac{cal}{g}$ = energy equivalent for protein

$9300 \frac{cal}{g}$ = energy equivalent for fat

Carbohydrate catabolized was calculated by subtracting energy obtained by protein catabolism from the total respired energy and dividing the result by 3500 cal/g.

Two assumptions are implicit in the above formulations: (1) All fat in larval tissues was synthesized rather than obtained in food. This is reasonable since the lipid content of food leaves is low, less than 1% (Schroeder, unpublished), and because the food passes rapidly through the gut (Wigglesworth, 1965). (2) All nitrogen in the feces resulted from protein catabolism rather than from non-assimilation. Since estimates of nitrogen assimilation range from 65% (Waldbauer, 1968) to over 91% (Crowell, 1941), this assumption is not entirely true. However, protein catabolism accounts for only about 2% of total water, therefore the error introduced is quite small. Both of these assumptions tend to cause underestimation of metabolic water with consequent overestimation of water efficiencies. The error would be in the

same direction and of the same order of magnitude for all individuals.

Budgets and Efficiencies

Dry Mass Budgets and Efficiencies

Calculations were made of individual dry mass budgets of the form:

$$I = G + R + E \qquad (6)$$

where: I = ingestion

G = growth

R = respiration

E = egestion

All factors were in dry mass units.

Dry weight ingested was estimated using live weight ingested and dry weight fractions of control leaves (I = live wt. x dry wt. fraction - residual). Dry weight of growth was calculated as the difference between initial dry weight (estimated at 15% of live weight) and terminal dry weight. Dry weight of feces was determined by direct weighing. Respiration was calculated by subtraction in Equation (6).

Gross (E^g) and net (E^n) conversion (utilization) efficiencies were calculated from Equations (7) and (8), respectively.

$$E^g = 100\ G/I \qquad (7)$$

$$E^n = 100\ G/(I-E) \qquad (8)$$

Assimilation efficiency was calculated as 100 times the ratio of E^g to E^n, or:

$$E^a = \frac{E^g}{E^n} = 100\ (I-E)/I \qquad (9)$$

Addition of an appropriate subscript denotes the type of utilization efficiency (m = mass, e = energy, n = nitrogen, and w = water).

Energy Budgets and Efficiencies

Prior to caloric determinations, dried leaves and feces were ground in a Wiley Intermediate Mill to pass through a size 40 mesh screen. Dried larvae were ground by hand in a mortar with pestle. All material was re-dried at 60°C over $CaCl_2$ in a partial vacuum for 24 hours before pelletizing with a Parr Pellet Press.

Caloric determinations were made on control leaves at three day intervals. Individual measurements were made for larval tissue, but feces were combined by species and instar. Whenever possible, there were three replicates of each determination.

When sample size permitted, calorimetry was done using a semimicro adiabatic bomb calorimeter (Parr Inst. Co.) requiring samples of approximately 0.2 g size. These were weighed using a Sartorius semimicro balance to a precision of 0.2 mg. For smaller samples (20 mg or less) the Phillipson micro bomb calorimeter (Gentry-Wiegert Inst. Co.) was used. These samples were weighed to a precision of 0.01 mg with a Cahn micro electrobalance.

Both calorimeters were standardized at every tenth run using benzoic acid pellets (Parr Inst. Co., 1960). Following each determination, bomb washings were titrated using 0.0375 N NaOH to obtain acid corrections (Paine, 1971). Energy equivalents were computer calculated.

Energy budgets and efficiencies were calculated by inserting caloric equivalents into the dry mass budget and efficiency equations (6), (7), (8), and (9).

Nitrogen Budgets and Efficiencies

Nitrogen percentages were obtained for the same material as selected for energy determinations. Powdered, dried samples appropriately sized to yield 400-700 μl gaseous nitrogen were weighed on the Cahn micro electrobalance and run on the Coleman Model 29 Nitrogen Analyzer. Percentage nitrogen was calculated by computer using instructions provided by Coleman Instruments (1968).

Nitrogen budgets and efficiencies were calculated using these percentages in the dry mass budget and efficiency equations (6), (7), and (9). Since nitrogen from catabolized protein appears in excreta rather than as respiratory gases, the net nitrogen efficiency, Equation (8) is trivial, any deviation from 100% resulting from measurement errors.

Water Budgets and Efficiencies

Water budgets in terms of weight of water were of the form:

$$I_w + M = G_w + Ev + E_w \quad (10)$$

where: I_w = weight of water ingested

M = weight of metabolic water produced

G_w = weight of water in growth

Ev = weight of water evaporated

E_w = weight of water egested

Calculation of gross and net efficiencies of water utilization were of the form:

$$E_w^g = 100\ G_w/(I_w+M) \quad (11)$$

$$E_w^n = 100\ G_w/(I_w+M-E_w) \quad (12)$$

Rates

Rates of growth (g), ingestion (i), excretion (f), assimilation (a), respiration (r), and production of metabolic water (m) were based on Gordon's (1968) mean exponential larval dry weight ($W_e = G/\ln(W_f/W_i)$), where W_f and W_i are final and initial dry weights, respectively. This gives a better estimate of mean weight than the arithmetic mean because growth is generally exponential. All rates were expressed in units of mg dry weight/(W_e x days).

Statistical Analyses

Statistical analyses including one-way analysis of variance, examination of differences among means, and partial correlations were carried out by computer using the Biomedical programs (Dixon, 1974). Simple linear regressions were done with the Stat-Basic Programs. Significance tests were made following instructions and using tables in Steel and Torrie (1960).

RESULTS

Table 1 gives means and standard errors for rates of growth, ingestion, assimilation, respiration and metabolic water production along with dry weight of the terminal larvae for each species and for 2 groups of Schiszura unicornis raised at different times. Regressions of all rates on dry weight were highly significant and negative (F-test, $p < 0.01$), as was regression of net water efficiency on dry weight (Table 2).

Table 3 gives means and standard errors by species of the conversion values which were applied to the dry mass budget equations for calculating energy and nitrogen budgets.

Individual gross and net efficiencies of dry mass, energy and water utilization and gross efficiency of nitrogen utilization along with species means and standard errors are given in Table 4. Larval concentration of energy, nitrogen and water relative to dry mass is demonstrated by the higher efficiencies for energy, nitrogen and water utilization relative to the equivalent dry mass efficiencies.

Analyses of variance, one-way classification, for differences among species and between two groups of S. unicornis raised at different times were significant for all rates, and for all efficiencies except nitrogen

utilization. The F-ratios with significance indicated are given in Table 5.

Because of the lack of significant interspecific differences in nitrogen utilization efficiencies, the total variance of E_n^g (37.5) was compared by an F-test to the total variance of E_e^n (91.1). As a result, variance in E_n^g was shown to be significantly less than variance in E_e^n ($F=2.43$, $p<0.05$).

The correlation matrix with significance indicated is given as Table 6. Gross energy efficiency shows significant positive correlations with gross nitrogen efficiency, leaf water, and rates of growth and water production, and a significant negative correlation with net water efficiency. However, net energy efficiency shows a significant correlation only with gross efficiency of nitrogen utilization (positive) and with rate of production of metabolic water (negative). In addition to its positive correlation with gross and net energy efficiencies, gross nitrogen efficiency is positively correlated with leaf water and growth rate, and negatively correlated with both net and gross water efficiency. Leaf water is positively correlated with gross efficiency of energy and nitrogen utilization and with rates of growth and water production. Leaf water is negatively correlated with both gross and net water efficiencies.

TABLE 1

MEANS AND STANDARD ERRORS BY SPECIES
DRY WEIGHT AND RATES OF GROWTH, INGESTION, ASSIMILATION, RESPIRATION AND METABOLIC WATER PRODUCTION

	dry wt.[a]	g[b]	i	a	r	m
Schiszura unicornis (J. E. Smith) (7/08-7/14)						
$\bar{x}$	87.1	398	2792	754	350	172
se	3.5	8	133	11	9	15
Hyalophora cecropia (8/16-9/02)						
$\bar{x}$	1923.6	66	564	126	60	27
se	71.8	2	4	1	1	3
S. unicornis (8/23-8/29)						
$\bar{x}$	86.6	186	1444	440	254	207
se	13.3	39	132	98	59	25
Simyra henrici (Grote & Robinson) (8/28-9/10)						
$\bar{x}$	111.9	202	1364	374	173	117
se	6.9	7	36	12	9	8
Hydria prunivorata (Ferguson) (9/05-9/09)						
$\bar{x}$	17.0	348	2186	956	609	394
se	0	42	238	134	92	62
Papillio glaucus (9/13-9/28)						
$\bar{x}$	222.6	159	1504	338	179	95
se	58.2	5	11	12	7	7

[a]Dry weights are expressed in mg.

[b]All rates are expressed in units of mg per g mean exponential dry weight per day.

TABLE 2

REGRESSIONS OF RATES OF GROWTH, INGESTION, ASSIMILATION, RESPIRATION AND METABOLIC WATER PRODUCTION AND WATER UTILIZATION EFFICIENCIES ON DRY WEIGHT OF TERMINAL LARVAE

Regression equation of the form: rate = a + b x dry wt.

	a	b	F
g	267.818	-0.110	7.774**
i	1903.571	-0.717	9.051**
a	577.858	-0.251	6.932**
r	308.718	-0.140	5.073**
m	196.244	-0.095	5.840**
E_w^g	42.033	0.002	0.035^{ns}
E_w^n	69.068	-0.012	4.897**

^{ns}Not significant

**$p < 0.01$

degrees of freedom for all tests = 5/15.

TABLE 3

ENERGY AND NITROGEN CONVERSION VALUES

	cal/g			%N		
	Larvae	Leaves	Feces	Larvae	Leaves	Feces
Schiszura unicornis (7/08-7/14)						
$\bar{x}$	6026	4562	4520	8.41	2.42	1.53
se	172	28	50	0.11	0.09	0.47
Hyalophora cecropia (8/16-9/02)						
$\bar{x}$	5965	4856	4724	9.14	2.78	1.96
se	67	51	17	0.28	0.17	0.42
S. unicornis (8/23-8/29)						
$\bar{x}$	5955	4860	4470	9.12	2.96	2.44
se	61	30	7	0.58	0.09	0.37
Simyra henrici (8/28-9/10)						
$\bar{x}$	6336	5026	4469	7.04	2.86	2.48
se	72	30	17	0.22	0.08	0.10
Hydria prunivorata (9/05-9/09)						
$\bar{x}$	6184	5006	4762	8.42	3.34	2.82
se	125	41	24	0.08	0.08	0.17
Papillio glaucus (9/13-9/28)						
$\bar{x}$	5650	4778	4573	7.63	2.51	2.10
se	137	68	34	0.10	0.17	0.22

TABLE 4

EFFICIENCIES OF UTILIZATION

	Dry Mass		Energy		Nit.	Water	
	E^g	E^n	E^g	E^n	E^g	E^g	E^n
Schiszura unicornis (7/08-7/14)							
$\bar{x}$	14.5	53.5	18.8	64.4	50.3	38.1	62.3
se	0.3	1.3	0.4	1.4	1.0	0.4	1.7
Hyalophora cecropia (8/16-9/02)							
$\bar{x}$	11.7	52.2	14.4	59.0	38.4	42.1	90.0
se	0.4	1.0	0.4	1.0	1.2	2.2	3.6
S. unicornis (8/23-8/29)							
$\bar{x}$	12.6	42.6	14.9	42.0	35.4	32.5	71.6
se	1.6	0.7	1.9	0.8	2.5	3.5	4.3
Simyra henrici (8/28-9.10)							
$\bar{x}$	14.8	53.8	18.3	56.7	39.5	48.7	81.2
se	0.7	1.6	0.9	2.5	0.5	1.6	2.3
Hydria prunivorata (9/05-9/09)							
$\bar{x}$	15.8	36.4	19.6	42.0	40.2	38.6	46.0
se	0.2	0.8	0.2	0.8	0.4	2.2	2.9
Papillio glaucus (9/13-9/28)							
$\bar{x}$	10.6	47.0	12.5	48.4	32.2	53.8	84.4
se	0.4	0.3	0.5	0.2	1.2	4.3	0.5

TABLE 5

F RATIOS AND SIGNIFICANCE FOR ANALYSIS OF VARIANCE AMONG SPECIES AND BETWEEN TWO GROUPS OF THE SAME SPECIES RAISED AT DIFFERENT TIMES

E_m^g	F = 4.0382*
E_m^n	F = 26.5413**
E_e^g	F = 25.1286**
E_n^g	F = 0.5586^{ns}
E_w^n	F = 5.8750**
g	F = 26.9256**
i	F = 43.0690**
a	F = 21.0790**
r	F = 19.8417**
m	F = 24.6138**

^{ns}Not significant

* $p < 0.05$

** $p < 0.01$

degrees of freedom for all tests = 5/15

TABLE 6

CORRELATION MATRIX

	E_e^g	E_e^n	E_n^g	E_w^g	E_w^n	Fec. H_2O	Leaf H_2O	g	m
E_e^g	1.000	0.309	0.697**	-0.149	-0.578*	-0.467	0.477*	0.717**	0.480*
E_e^n		1.000	0.645**	0.259	0.262	0.166	-0.137	0.194	-0.544*
E_n^g			1.000	-0.614*	-0.523*	-0.116	0.621**	0.758**	0.161
E_w^g				1.000	0.574*	0.134	-0.389	-0.307	-0.498*
E_w^n					1.000	0.493*	-0.764*	-0.811**	-0.913**
Fec. H_2O						1.000	-0.094	-0.271	-0.550*
Leaf H_2O							1.000	0.805**	0.641**
g								1.000	0.646**
m									1.000

* $p < 0.05$

** $p < 0.01$

all others are non-significant

DTSCUSSION

The highly significant negative regressions of rates of growth, ingestion, assimilation, respiration and metabolic water production on dry weight confirms the inverse relationship between size and weight specific metabolism (Gordon, 1972). The negative regression of net water efficiency on dry weight may be due partly to the higher respiration rate of smaller individuals resulting in high rates of water exchange across respiratory membranes. This may also be due in part to the greater surface to volume ratios of smaller individuals.

Gross efficiency of nitrogen utilization is more than triple the corresponding dry mass efficiency and more than twice the corresponding energy efficiency. This may be indicative of the differential accumulation of nitrogen. while energy increases about 30-40% from leaf to larval tissue, nitrogen density increases about 200-300%. Greater accumulation of nitrogen could result either from increased efficiency of extraction of nitrogen or from increased ingestion of dry mass. Either of these would result in greater nitrogen efficiency relative to dry matter or energy. A similar argument holds for the increase of energy density from leaf to larval tissue, and for the increase in water concentration from leaf to larval tissue.

A comparison from Table 5 of the highly significant interspecific differences of net energy efficiency with the lack of significant differences in gross nitrogen efficiency suggests that interspecific differences may be limiting to energy efficiency, while nitrogen efficiency may be limited by external constraints more universally applicable to the life strategies of these species. Since all larvae were feeding on leaves of approximately the same caloric value and increasing the energy density to their own tissues by varying amounts (observed values for larval tissue varied from 5650 cal/g to 6356 cal/g) some of the efficiency differences may be accounted for by the metabolic cost for increasing energy density from leaf to larval tissue (see Schroeder, 1977_b). The need to concentrate specific nutrients, i.e. nitrogen, or water would also have an effect on energy efficiency because those larvae that are better at concentrating the nutrient may not waste as much energy in obtaining their requirements. At some point, an energy trade-off occurs where the metabolic energy cost of concentration exceeds the energy that would be wasted in increasing ingestion. The lack of variability in the nitrogen efficiency data may lend support to this interpretation of differences in limiting factors.

Since net nitrogen utilization efficiency approaches 100%, gross nitrogen utilization efficiency approaches nitrogen assimilation efficiency as a limit. Therefore any

mechanism which limits nitrogen assimilation would be also limiting to nitrogen utilization efficiency. Schroeder (1977_c) has proposed that cyanogenic compounds in Prunus serotina leaves may limit nitrogen (protein) assimilation by poisoning the active transport mechanisms in the larval gut (Jones, 1972). Rhodanese is an enzyme which detoxifies cyanide (Westley, 1973). If cyanide is important in limiting nitrogen assimilation, those larvae with high levels of rhodanese in the gut would be expected to have higher assimilation and utilization efficiencies for nitrogen. Without measuring both leaf cyanide and larval gut rhodanese, it is impossible to tell if this mechanism is operating to limit nitrogen utilization.

In spite of its high variability, net energy efficiency is correlated only with gross nitrogen efficiency and with production of metabolic water (Table 4). These data do not support Scriber's (1977) study showing a direct correlation between energy efficiency and water availability. Since Scriber used only one species in his study, the presence here of significant interspecific differences might mask small differences due to water stress. Additionally, species may be adapted to the level of water availability they normally encounter during their feeding period so that energy efficiencies do not correlate with water availability. Some of the food leaves used by Scriber were water supplemented with the result that he observed higher water levels ($> 75\%$) than were observed here (50-65% of fresh leaf weight).

The limiting effect of water availability on nitrogen efficiency seems, however, to be quite pronounced. Not only is there a high positive correlation between leaf water and nitrogen efficiency, there is a high negative correlation between water efficiency and nitrogen efficiency. This may lend credence to the argument that nitrogen efficiency is maximized since this is what seems to be most seriously affected by water stress.

A possible synergism between nitrogen efficiency and leaf water is evidenced by the high positive correlation with both of these and rate of growth. Two explanations are possible here. It may be that high leaf water leads to high nitrogen efficiency, and this rapid accumulation of nitrogen accelerates growth rate. On the other hand, if high water availability directly accelerates growth rate, the rapid growth may allow for more efficient accumulation of nitrogen. This is contrary to Slansky and Feeny (1977) where nitrogen accumulation and growth rates were stabilized over a wide range of nitrogen availability. It also appears to be contrary to Odum and Pinkerton's (1955) hypothesis of rate maximization at relatively low efficiencies. However, since a graph of efficiency against rate in the Odum-Pinkerton hypothesis would yield a bell shaped curve, the observed relationship for any narrow range of efficiencies and rates depends on their position on the curve. If the observed values lie on the increasing portion of the curve, a positive correlation is expected.

The differences between energy and nitrogen efficiencies with respect to correlations is difficult to explain in terms of evolutionary strategies. It may be that energy efficiency is not maximized and is, therefore, little affected by the slight variations found in the environment. On the other hand, if energy is of overwhelming importance, evolution may have resulted in species adaptations which allow maximization of energy efficiency in spite of the environmental differences generally encountered. This second interpretation may be favored by the fact that when leaf water (Scriber, 1977) or leaf nitrogen (Slansky and Feeny, (1977) are artificially manipulated outside of the usual environmental levels, energy efficiency responds.

SUMMARY

In field collected lepidopteran larvae, water efficiency and rates of growth, ingestion, assimilation, respiration and metabolic water production were found to vary inversely with dry weight of the terminal larva. This is in agreement with the generally held theories of reduction of weight specific metabolism with increase in size and of the importance of surface-volume ratios in water economy.

Energy efficiency was found to have highly significant interspecific differences and high variability, while nitrogen efficiency had non-significant interspecific differences and low variability. The interpretation proposed was that limits to energy efficiency were somewhat dependent on interspecfic differences while limits to nitrogen efficiency were external and universally applicable according to life strategy.

Energy efficiency showed few correlations with investigated parameters while nitrogen efficiency showed many. This lends support to the suggestion that energy is internally limited and nitrogen is externally limited. It was also suggested that energy efficiency may be of such importance that adaptation to environmental conditions may result in its being little affected by minor variations in such conditions.

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