

ORGAN GROWTH AND OXYGEN CONSUMPTION IN LAYSAN ALBATROSS EMBRYOS

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ABSTRACT.—Embryonic oxygen consumption and organ growth were measured in the Laysan Albatross (*Diomedea immutabilis*). The logarithms of oxygen consumption and organ mass increased linearly with the logarithm of whole-embryo wet mass. The rate of growth of skeletal muscle and the gastro-intestinal tract exceeded that of the embryo as a whole, while the heart, lungs, brain, and eyes grew more slowly. The absence of a discernible effect of pipping of the egg on oxygen consumption and organ growth in the embryonic albatross, in contrast to a previous report for the Wedge-tailed Shearwater (*Puffinus pacificus*), was attributed to a different sequence of events during pipping in the two species. Received 6 July 1992, accepted 15 April 1993.

In a previous paper Zhang and Whittow (1992) reported on embryonic oxygen consumption and organ growth in the Wedge-tailed Shearwater (*Puffinus pacificus*), a tropical, procellariiform seabird. Substantial differences in growth rates between different organs were observed in unpipped eggs, and further differences occurred after the eggs had pipped. This was the first study of embryonic organ growth in a procellariiform bird. In order to determine if the pattern of growth observed in the shearwater is characteristic of Procellariiformes in general, similar data were collected from the Laysan Albatross (*Diomedea immutabilis*). These data form the basis of the present report. The Laysan Albatross was chosen for investigation because the sequence of events during pipping of its egg differs from that in the Wedge-tailed Shearwater (Pettit et al. 1982).

MATERIALS AND METHODS

Laysan Albatross eggs were collected by U.S. Dept. of Agriculture (Animal Damage Control) personnel from various sites in the main Hawaiian islands. The eggs were transported in insulated containers and, in some instances, were incubated at 36°C while awaiting shipment to the laboratory. In the laboratory, the eggs were incubated at 36°C and 60% relative humidity in a commercial, forced-draft, incubator (GQF, model 1202) and turned at least twice daily. Data were collected from unpipped eggs, internally-pipped eggs, externally-pipped eggs with star-fractured shells, and eggs with pip holes (Pettit et al. 1982).

The oxygen consumption ($\dot{V}_{O_2}$) of eggs of different estimated ages was measured in a modified Scholander respirometer (Ackerman et al. 1980). The chamber containing the egg was immersed in a water bath at 36°C and the chamber was ventilated with air for 60 min before measurements began. The oxygen consumption was measured over a 60-min period by introducing measured volumes of oxygen into the chamber to bring the meniscus of the manometer between the egg chamber and its compensating chamber back to zero. Carbon

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dioxide produced by the embryo was absorbed by Ascarite placed at the bottom of the chamber. All values of oxygen consumption were corrected to STPD.

After the oxygen consumption had been determined, the egg was opened and the embryo carefully separated from the yolk sac. The yolk-free wet mass of the embryo was determined on a Mettler balance (model H6) to the nearest 0.1 mg. The following linear dimensions of the embryo were then measured with a dial caliper: (1) whole embryo length—top of the head to the tip of the tail, (2) culmen length—tip of the beak to the beginning of the nasal skin on the upper mandible, (3) wing length—tip of the wing to the shoulder joint, with the wing extended, (4) leg length—tip of the middle toe to the head of the femur, (5) toe length—tip of the nail to the proximal fold in the webbing between the middle and lateral toe, and (6) neck length—from the lower mandible to the thirteenth cervical vertebra.

The embryo was then carefully dissected, and the following organs and tissues were weighed, wet, on the Mettler balance: (1) leg muscles—muscles from both legs, (2) pectoral muscles—muscles from both sides, (3) heart, (4) liver, (5) stomach, (6) intestine, (7) lungs, (8) brain, and (9) eyes.

As the age of the embryos was not known exactly, whereas the embryonic mass could be measured precisely, all measured data were related to embryo mass. However, a good estimate of the embryonic age can be derived, if needed, from previously published data (Pettit et al. 1982).

The basic statistical procedure used was to plot the logarithm of the $\dot{V}_{O_2}$ or organ mass against the logarithm of the body mass and to compute a linear regression line, together with 95% confidence limits.

RESULTS

Oxygen consumption.—The data for oxygen consumption ($\dot{V}_{O_2}$; Fig. 1) reveal that the logarithm of oxygen consumption of the embryo increased linearly with log body mass ($r = 0.917$).

Organ mass.—Heart, lungs, and brain.—The mass of the heart, lungs and brain increased linearly with the increase in the mass of the whole embryo on a log-log scale (Fig. 2). The slopes of the relationships were all less than 1 (Table 1). *Eyeballs.*—The relationship between log eyeball mass and log embryo mass also yielded a slope that was less than one (Fig. 2, Table 1). *Leg and pectoral muscles.*—In contrast to the heart, lungs, brain, and eyeballs, the slope of the relationships between the log of the leg and pectoral muscles and log embryo mass exceeded unity (Fig. 2, Table 1). *Stomach, intestine, and liver.*—The stomach, liver and intestine all increased linearly with embryo mass (Fig. 3) on a log-log plot, and the slopes of the relationships exceeded unity (Table 1). *Linear dimensions.*—Although the overall length of the embryo increased linearly with body mass on a log-log plot (Fig. 4), there was a trend, particularly evident in neck and wing length, for the length of body parts to increase little after pipping.

DISCUSSION

Embryonic oxygen consumption and organ mass of the Laysan Albatross increased linearly with whole-embryo mass on a log-log plot. There

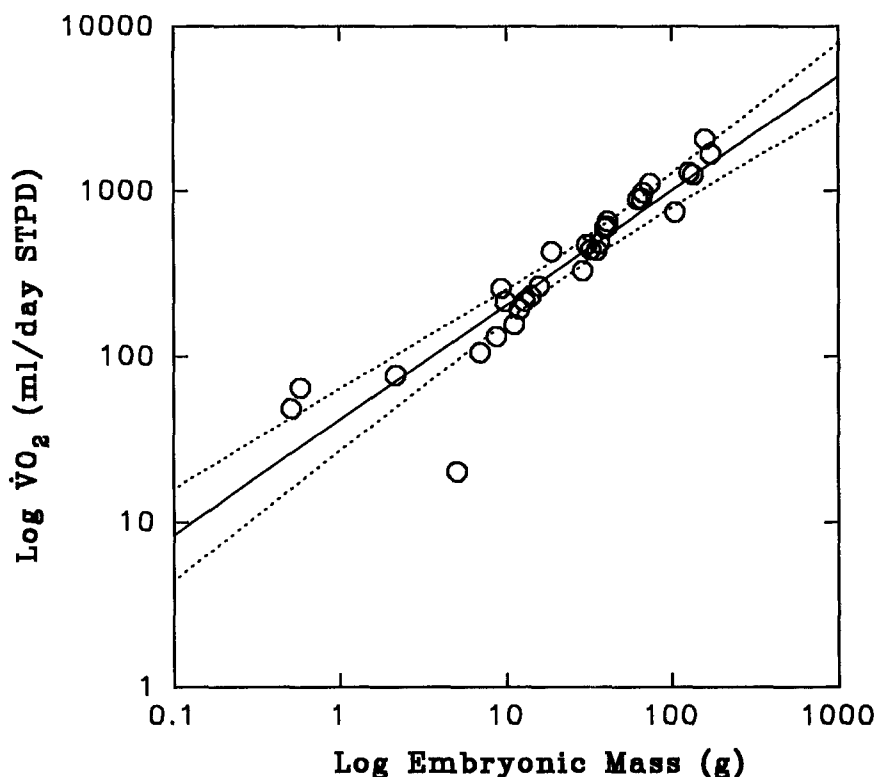


FIG. 1. Oxygen consumption ($\dot{V}_{O_2}$) of Albatross embryos of known body mass. The solid line is the fitted linear regression of $\log \dot{V}_{O_2}$ on $\log$ embryo mass. The dashed lines define the 95% confidence limits. Intercept and slope of the regression line are 1.616 and 0.694, respectively.

was no discernible effect of pipping (except, possibly, in the linear dimensions which tended to increase little in pipped eggs, Fig. 4). There were, however, differences in the slopes of the relationships for different organs (Fig. 5). The steepest slope was for the leg muscles while the log relationship between the eyeball mass and whole-embryo mass had the lowest slope (Fig. 5). The slopes of the regressions of muscle and the gastro-intestinal tract exceeded unity, indicating that those tissues were growing faster than the embryo as a whole. The heart, lungs, brain, and eyeballs, on the other hand, were growing more slowly.

It is difficult to compare the results of the present investigation with those reported earlier for the Wedge-tailed Shearwater (Zhang and Whitrow 1992), because the data are presented differently in the two studies. However, a striking feature of the relationships in the shearwater, regardless of the way in which the data are analysed, is the acceleration of

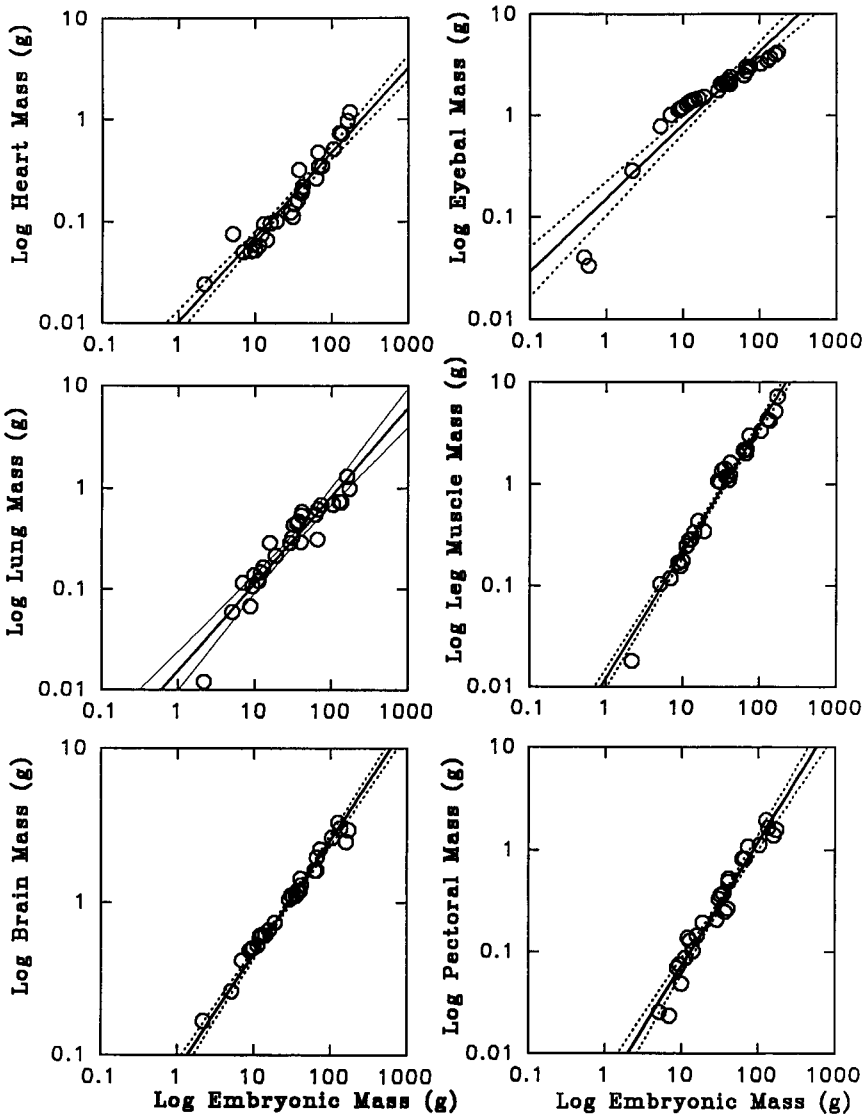


FIG. 2. Organ wet mass as a function of wet whole-embryo mass in the Laysan albatross. A linear regression line is fitted to the log, log, data. Notations as in Fig. 1. Intercepts, slopes, and correlation coefficients are given in Table 1.

TABLE 1
INTERCEPTS, SLOPES AND CORRELATION COEFFICIENTS FOR THE LINEAR REGRESSION OF LOG
ORGAN MASS ON LOG EMBRYO MASS FOR LAYSAN ALBATROSS EMBRYOS

Organ	Intercept	Slope	r ^a
Brain	-1.099	0.752	0.991
Heart	-1.983	0.828	0.976
Lungs	-1.809	0.863	0.947
Eyeballs	-0.818	0.722	0.939
Leg muscles	-1.922	1.246	0.993
Pectoral muscles	-2.368	1.221	0.976
Intestine	-2.153	1.116	0.971
Liver	-1.992	1.069	0.981
Stomach	-1.911	1.242	0.984

^a Correlation coefficient.

oxygen consumption in pipped eggs. The increased oxygen consumption was associated with an increased slope of the mass of the intestine on whole-embryo mass. Neither of these phenomena was apparent in the albatross data. The most likely explanation for these differences between the two species would appear to be the different sequence of events during pipping in the shearwater and albatross. In the shearwater, the initial event during pipping was a series of star-fractures of the shell (external pipping), which results in an increase in the gas conductance of the shell (Pettit and Whittow 1983) and an increase in the partial pressure of oxygen (P_{O_2}) in the aircell gas (Pettit and Whittow 1982) allowing the embryo to increase its oxygen consumption. In the Laysan Albatross, the initial event was internal pipping in which the embryo penetrates the aircell of the egg with its beak (Pettit et al. 1982), leaving the gas conductance of the shell unchanged. Internal pipping permits ventilation of the lungs with aircell gas, leading to a substantial reduction in the P_{O_2} and a rise in the carbon dioxide (P_{CO_2}) of aircell gas (Pettit et al. 1982). Thus, an accelerated increase in embryonic oxygen consumption in the Laysan Albatross in the early stage of pipping could be achieved only at the expense of exposure of the embryo to hypoxia and hypercapnia. This may be an explanation for the absence of such an increase in the albatross. It is more difficult to ascribe the intestinal growth in pipped shearwater eggs except to state that intestinal growth and oxygen consumption are related. It might have been expected that the relatively early development of pulmonary function in the albatross would be associated with a high rate of growth of the lungs before pipping, but such was not the case. It is possible that increased lung maturation rather than accelerated growth occurs during this period.

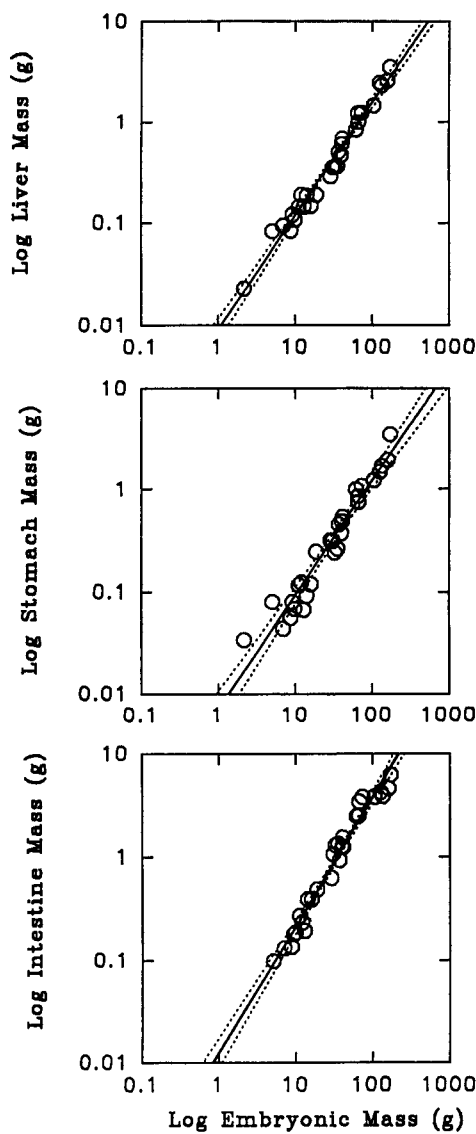


FIG. 3. Wet mass of the stomach, intestine and liver as a function of wet whole-embryo mass in the Laysan Albatross. Notations as in Fig. 2.

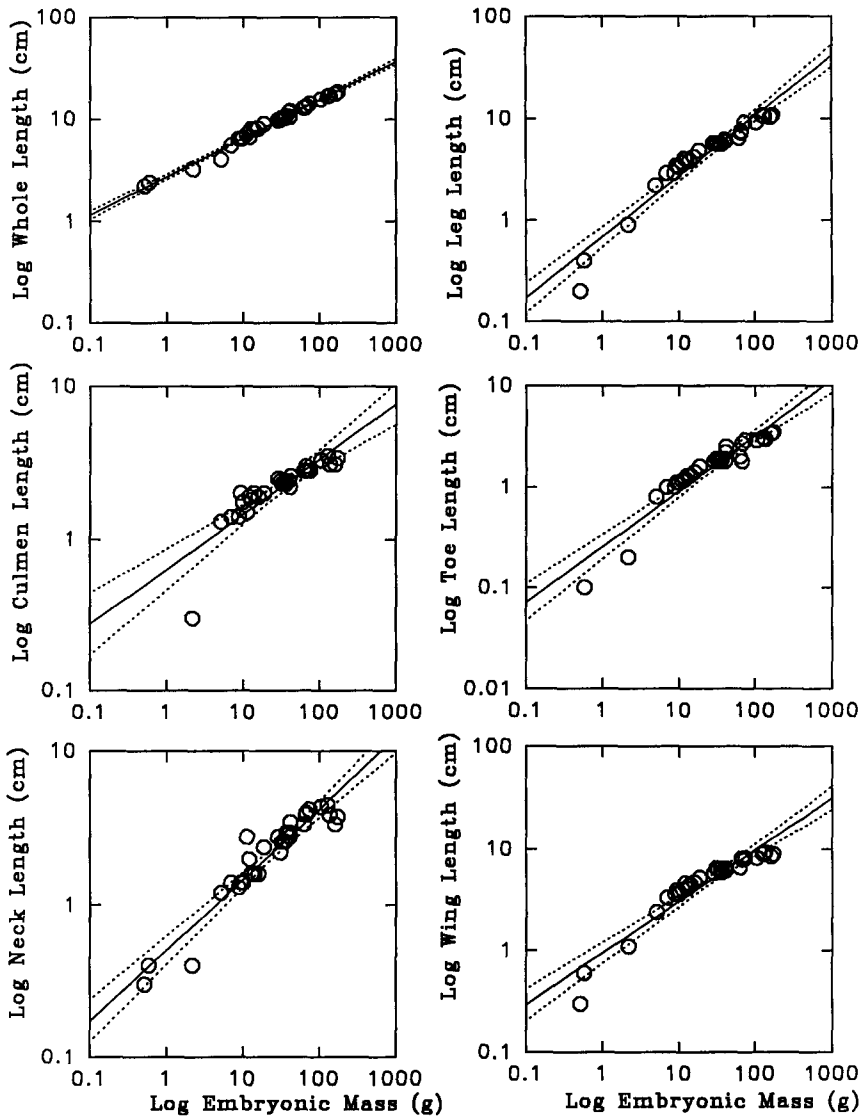


FIG. 4. Embryo lengths in relation to wet embryo mass. Notations as in Fig. 1.

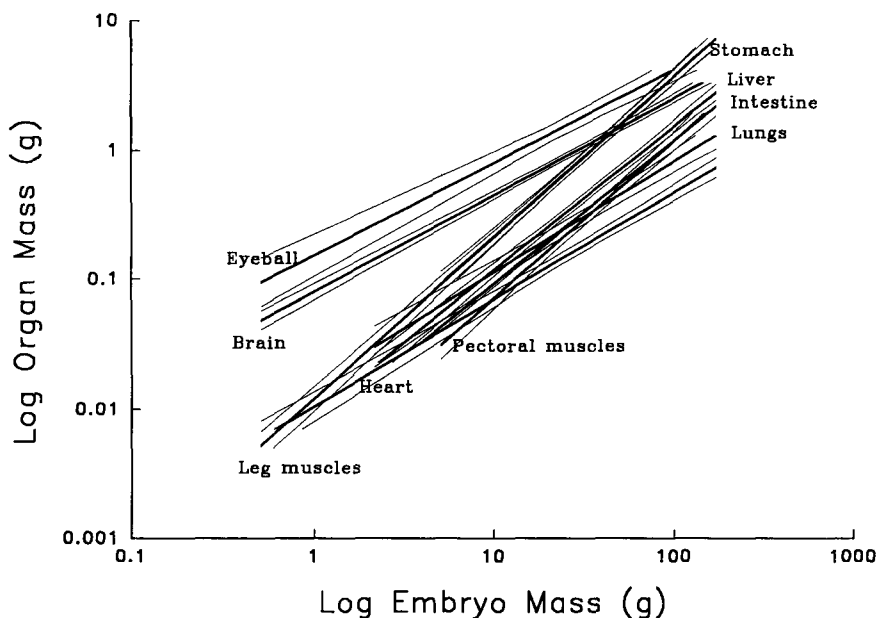


FIG. 5. Linear regression lines and confidence limits of organ mass on embryo mass.

The results of the present investigation correlate well with those of a parallel study (Tazawa and Whittow, unpubl. data) in which embryonic heart rate and oxygen pulse were measured in the Laysan Albatross and Wedge-tailed Shearwater. In that study, heart rate and oxygen pulse accelerated during pipping in the shearwater but not in the albatross. In the shearwater, but not in the albatross, cardiac mass increased faster than that of the embryo as a whole. Consequently, by the time that pipping occurred, the heart was relatively larger in the shearwater than in the albatross.

In conclusion, it would seem that there is not a procellariiform pattern of embryonic organ growth and oxygen consumption but that these parameters, like the sequence of events during pipping, vary among different species.

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