

Modification of Optic Stowaway Tidbits Used for Long Term Measurement of Body Temperature in Animals

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Abstract: The Optic Stowaway Tidbit is a commercially available temperature-sensitive data logger (dimensions: 2.6x2.6x0.7 cm, weight: 8.7 g). Although it was manufactured for climatological and industrial applications, it is successively used for monitoring long-term body temperatures in free-ranging and laboratory animals. The data logger has an expected life span of 5 years because of the limited life of its non-replaceable battery. This paper describes how Optic Stowaway Tidbits can be modified without compromising the function of the data loggers. Weighing around 15 g after potting with wax, the data loggers can be implanted intraperitoneally into animals larger than 300 g. Calibration data showed that the uncalibrated data loggers can lead to errors in the recorded temperatures, confirming that Optic Stowaway Tidbits must be calibrated prior to implantation.

Key words: Calibration, data logger, thermoregulation, Wistar albino rat

Introduction

In thermoregulatory studies, it is fundamental to obtain body temperature (T_b) recordings from undisturbed, unstressed animals. However, it is not possible to record body temperature reliably with conventional measurement techniques, which require handling, restraining, and presence of investigator nearby the animal, because these stress-inducing factors lead to several physiological changes (e.g. alterations in core and skin temperature; POOLE, STEPHENSON 1977, BERRY *et al.* 1984, LONG *et al.* 1990, CABANAC, BRIESE 1992, GORDON *et al.* 2002). With the advance of electronic and computer technology, numerous monitoring systems (e.g. biotelemetry, data logging), which permit long-term continuous recording of body temperature from freely moving animals in their home cages or own habitat without disturbing them, have been developed (ANDREWS 1998, KRAMER, KINTER 2003). These systems are free from stress-inducing factors associated with conventional measurement

techniques, which confound the interpretation of data (MALKINSON, PITTMAN 1997).

Biotelemetry is successfully used in many and diverse areas of animal research (KRAMER, KINTER 2003). However, telemetry technology may not be appropriate in certain conditions. Short transmission range of telemeters often limits their application in free-ranging animals in several kinds of environments (e.g. dense forests, urban settings, oceans; ANDREWS 1998). Also, short battery life of telemeters prevents their use in studies which require long-term continuous recordings of body temperature (e.g. hibernation studies; KAMERMAN *et al.* 2001). In such situations, if free-ranging animals can be easily recaptured, data loggers may offer some advantages over telemetric devices: (1) data is stored on the board memory for later retrieval, requiring no receiving equipments or persons on-site for data obtaining (KAMERMAN *et al.* 2001, LOVEGROVE 2009) and (2) they have high sam-

pling rate, multiple sampling options, and long battery life (up to 10 years).

The recent availability of miniature temperature-sensitive data loggers (e.g. Thermochron ibutton, Optic Stowaway Tidbit) has made collecting body temperature of the animals much easier (BOYLES 2007). For example, Optic StowAway Tidbit (Model TBI 32, Onset Computer Corporation, Pocasset, Mass., USA) can store large numbers of time and date stamped data points (32520, configurable from 0.5 sec to 9 h time intervals with a resolution of 0.16 °C), have high accuracy (0.2 °C), are easy to program and have an acceptable size for implantation into the medium-sized animals. Therefore, they are commonly used in biological studies, which especially require long-term and/or high-resolution temperature data (FULLER *et al.* 1999, PULAWA, FLORANT 2000, KAMERMAN *et al.* 2001, HUT *et al.* 2002, MAC LEAY *et al.* 2003, LEHMER, BIGGINS 2005, NICOL, ANDERSEN 2006, LONG *et al.* 2007, KART GÜR *et al.* 2009, LEE *et al.* 2009, HARLOW *et al.* 2010, VON BRANDIS *et al.* 2010). The logger's expected life span is 5 years because of the limited life of its non-replaceable battery.

Here, to make the battery user-replaceable, I described a simple and inexpensive method of modification of Optic StowAway Tidbit, which I previously used to record body temperature of free-ranging, hibernating Anatolian ground squirrels, *Spermophilus xanthorpymnus* (KART GÜR 2008, KART GÜR *et al.* 2009). I also evaluated the performance of the modified data loggers by recording the body temperature of laboratory rats over several days under the controlled laboratory conditions.

Material and Methods

Replacing the battery

The Optic StowAway Tidbit (Model TBI 32) comprises a printed circuit board, which integrates a thermometer, real time clock, and non-volatile memory (EEPROM) for storing temperature, time, and date recordings, and a non-replaceable 3V lithium battery. The PCB has been soldered to the battery with solder tabs attached (Fig. 1a). In order to remove the old battery, two solder points on the PCB (Fig. 1b) should be de-soldered using a fine tip soldering iron. The battery polarity should be noted down before attempting to remove the old battery. The new battery

(Model CR 2450 N) should not be soldered directly to the PCB. This procedure may damage both the PCB and new battery. Thus, I recommend the use of new batteries with solder tabs attached, which are available upon request from most major battery suppliers. Two pieces of electrically identical wires (e.g. CAT 5 cables) need to be used to provide a contact between the PCB and new battery. One end of each wire should be soldered to the new battery and the other end to the corresponding point on the PCB to which the old battery was soldered (Fig. 1c). As a final step, the new battery should be wrapped around with a piece of electrical tape to prevent shorting out. After replacing the old battery, it is important to check the new battery voltage with a voltmeter. It should be 3 volts.

Waxing

After confirming that the communication of the modified Optic StowAway Tidbits with Optic Base Station is working, each has been programmed to log Tb every minute during the experiment. Before waxing, the data loggers were wrapped around with a piece of cling wrap and parafilm (Fig. 2). This procedure prevents wax from sticking to the PCB and also makes the data loggers suitable for uniform waxing. To improve water resistance and prevent tissue reaction in the animal, the data loggers were coated in paraffin-eltax compound (Minimitter Company, Oregon, USA, <http://www.minimitter.com>) and then were cold-sterilized in glutaraldehyde solution for 24 h before being implanted into the peritoneal cavity of laboratory rats.

Calibration

Before and after the experiment, the wax coated modified Optic StowAway Tidbits were calibrated in an insulated water bath (MGW Lauda C6, Westbury, NY) at water bath temperatures of from 33 to 41 °C using a calibrated and certified mercury thermometer (with a range of : -1 to 50 °C and a resolution of 0.1 °C, Allafrance, UMS, Ankara, Turkey). The data loggers were maintained at five calibration steps for 30 min each. The temperature of the water bath was kept constant for 30 min before each calibration step. Only stable (plateau) temperature measurements were included in the analyses. For each data logger, linear regression model was fitted to the calibration data and therefore slope, intercept, and r^2 were calculated.

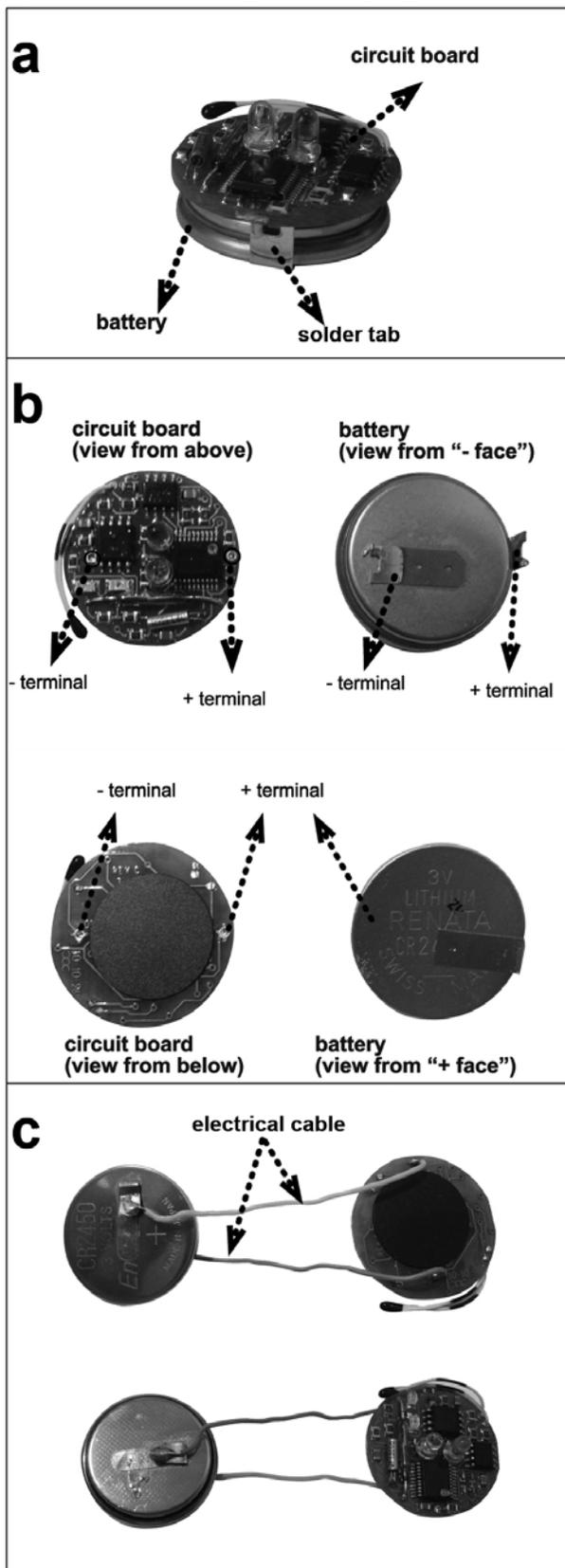


Fig. 1. (a) An intact Optic StowAway Tidbit. (b) The circuit board detached from the battery. (c) The modified Optic StowAway Tidbit ready for being wrapping and waxing.

Animals

To evaluate the function of the modified Optic StowAway Tidbits, five male Wistar albino rats (from a breeding colony at Department of Pharmacology and Clinical Pharmacology, Ankara University) were used. Body weights ranged from 305–350 g. The animals were housed individually in macrolon cages lined with wood shavings and were maintained at environmental temperature (T_a) of 21 ± 1 °C and humidity of 40% under a 12L:12D cycle with lights on from 07:00-19:00. Rat pellets (Bilyem, Ankara) and tap water were available ad libitum. All animal care and use protocols were consistent with international guidelines for care and use of laboratory animals (ILAR 1996).

Implantation

The wax coated, modified Optic StowAway Tidbits were surgically implanted into the peritoneal cavity of male Wistar albino rats under general anesthesia and sterile conditions. The animals were anesthetized with intraperitoneal injection of ketamine (80 mg/kg, Ketazol® 10%, Richter Pharma AG, Wels, Austria) and xylazine (10 mg/kg, Alfazyne® 2%, Alfasan International B. V. Woerden, Netherlands). The lower part of the abdomen was shaved and scrubbed with povidone-iodine solution (Isosol, 10%, Merkez Lab., Istanbul). A 1.5-2 cm midline incision was made in the skin and *linea alba* in the ventrum beginning approximately 2 cm above the genital. The data logger was then inserted into the peritoneal cavity of the animals. The abdominal muscles and skin were closed separately with absorbable (chromic catgut, 3/0) and non-absorbable (silk, 4/0) sutures in a simple interrupted pattern, respectively. The data loggers were recovered two weeks after implantation. Any macroscopic sign of injury or inflammation caused by the data loggers was checked at the time of retrieval.

Body temperature recording

T_b of male Wistar albino rats was recorded continuously at 1-min intervals throughout 15 days of the experiment by the modified Optic StowAway Tidbits. The measurement range of the data loggers was -5 to +44 °C, which is not standard, but custom made. The data loggers were launched and downloaded (BoxCar Pro 4.3, Onset Computer Corporation, USA) using infrared communication between the data logger and a base station (Optic Base Station, Onset Computer



Fig. 2. Preparation of an Optic StowAway Tidbit before being implanted into an animal.

Corporation, USA), which was linked to the computer via USB or serial port.

Results and Discussion

The methods of modifying Optic StowAway Tidbits described here showed that the battery of the data loggers is easily, conveniently, and inexpensively user-replaceable. The overall cost of supplies necessary for the modification (e.g. battery, elvax, electrical cable) is low, approximately 2 USD per animal. This cost constitutes only 1.3% of the cost of a new, intact Optic StowAway Tidbit (159 American dollars at the time of writing of this paper; www.onset.com).

The weight of the unmodified Optic StowAway Tidbit was 8.7 g as supplied. The modification increased the weight of the data logger to 10.4 ± 0.9 g (mean \pm SD, range = 10.2-10.4, $n = 5$), which constitutes 120% of the weight of the unmodified data logger. The weight of the wax coated, modified data logger was 14.65 ± 0.94 g (range = 13.85-14.81, $n = 5$). Although the final weight of the data loggers corresponded to 3-5% of the total body weight guideline recommended by MACDONALD, AMLANER (1980), they appear to be large for implantation into the peritoneal cavity of the laboratory rats. Some investigators reported that large implants may cause severe surgical complications (e.g. seroma formation, skin lesions in rats; MORAN *et al.* 1998). In this study, no macroscopic sign of injury or inflammation associated with the data logger was observed in the peritoneal cavity of the animals at the time of retrieval surgeries. Therefore, the sizes of the modified data loggers seemed suitable for laboratory rats of the body sizes represented in this study (305-350 g).

The uncalibrated data from the modified Optic StowAway Tidbits indicated a spread of about 0.85 °C and a parallel response to rapid fluctuations in water bath temperature (Fig. 3a). The r^2 of the calibration curves of the data loggers calibrated both before and after the experiment was 0.999, suggesting that about 100% of the variation in the temperatures recorded by the thermometer was explained by the data loggers. The slope of the calibration curves of the data loggers before and after the experiment was 0.999 ± 0.015 °C (Mean \pm SD, range = 0.973-1.020 °C, $n = 5$) and 1.002 ± 0.019 °C (range = 0.974-1.016 °C, $n = 5$), respectively, and the intercept 0.276 ± 0.473 °C (range = -0.454-0.866 °C, $n = 5$) and 0.086 ± 0.636 °C (range = -0.686-0.839 °C, $n = 5$), respectively. There appeared to be little calibration drift throughout the experiment. A typical calibration curve obtained both before and after the experiment was shown in Fig. 3b.

The calibration data confirmed that Optic StowAway Tidbits should be calibrated prior to implantation. That data loggers were not calibrated can lead to errors as much as about 1.0 °C in the recorded temperatures. Thus, calibration is very essential especially for studies where Tb must be recorded very accurately (e.g. studies on fever or temperature-dependent age determination). To assure that data loggers have not drifted from the initial calibration, they should also be calibrated, at least, both before and after the experiment.

All of the modified Optic StowAway Tidbits provided high quality recordings of the body temperature of laboratory rats throughout the experiment (Fig. 4), suggesting that the modified data loggers retained the functionality of the original ones

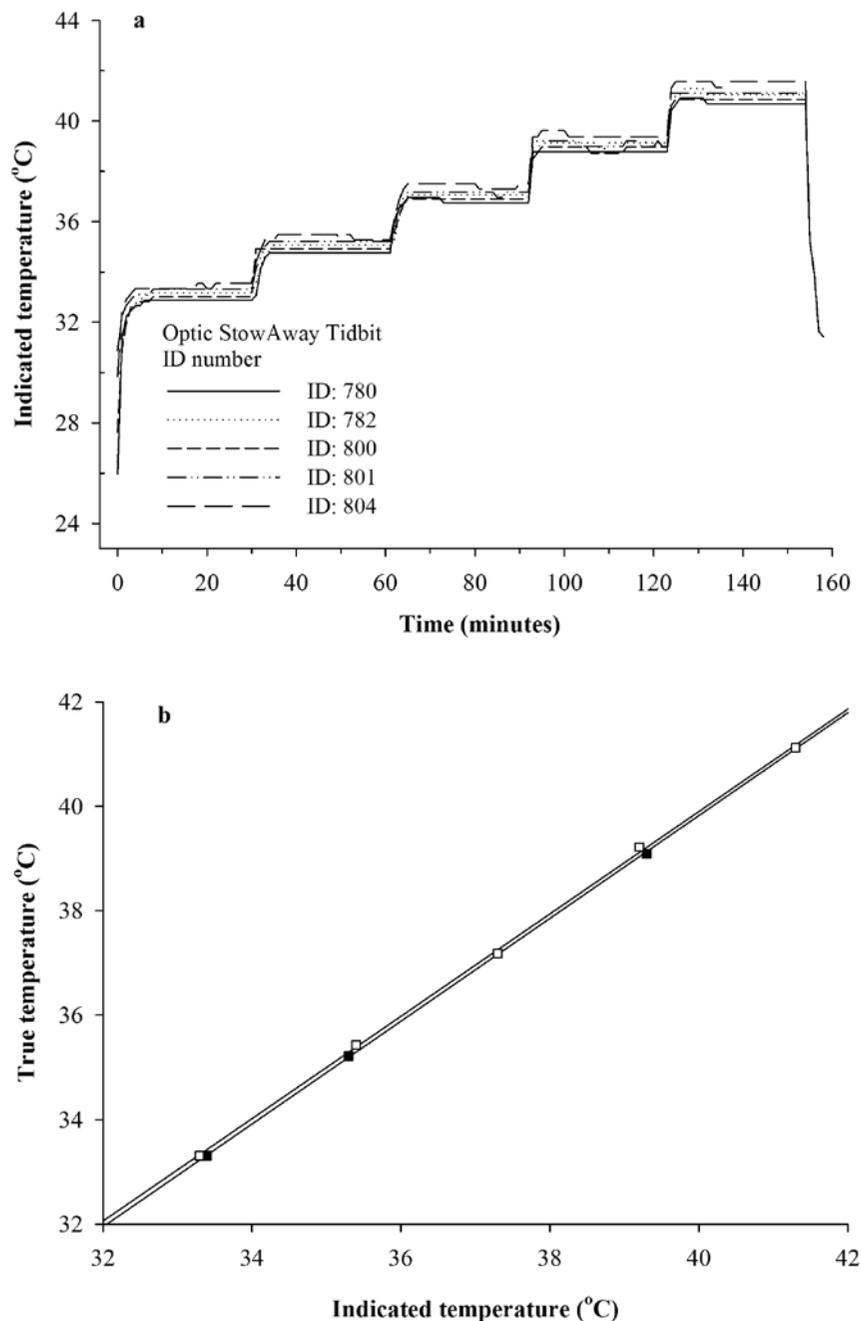


Fig. 3. (a) The uncalibrated data from the modified Optic StowAway Tidbits at water bath temperatures of from 33 to 41°C. (b) The calibration equation of a modified Optic StowAway Tidbit before (solid squares: $y = -0.454 + 1.016x$, $r^2 = 1.000$; $S_{y,x} = 0.005^\circ\text{C}$) and after (open squares: $y = -0.686 + 1.020x$, $r^2 = 0.999$; $S_{y,x} = 0.014^\circ\text{C}$) the experiment.

throughout the experiment. However, it should be remember that being inside an animal for a long period of time may cause some problems to the integrity and/or functionality of the modified data loggers.

The body temperature patterns of the animals were similar to those of other nocturnal rodents, including laboratory rats, with low values during the light phase of the light-dark cycle and high values during the dark phase (Fig. 4, REFINETTI, MENAKER

1992, AKARSU, MAMUK 2007). The animals also exhibited a slight rise in body temperature in the morning (around 10:00 a.m., Fig. 5). It was likely due to that the animals were disturbed while the laboratory was cleaned each day at around 10:00 a.m. (E. S. AKARSU, personal communication 2010). It is well known that some procedures (e.g. noise, handling, weighing, blood sampling) causes a rapid rise in body temperature, often called 'stress induced hyperthermia' in rats (LONG *et al.* 1990).

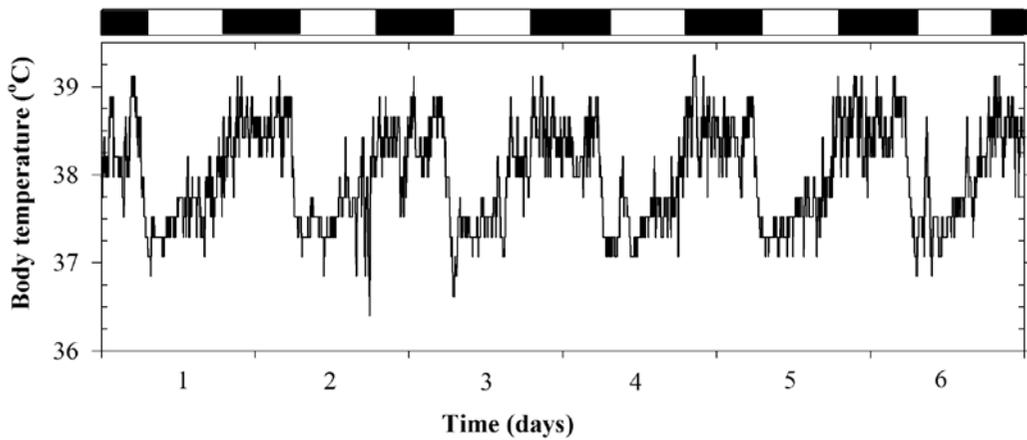


Fig. 4. Six day section of the records of body temperature of a male Wistar albino rat maintained under a 12:12h light-dark cycle. Lights were on daily from 07:00 to 19:00 h. Data were collected and plotted in 1 min interval.

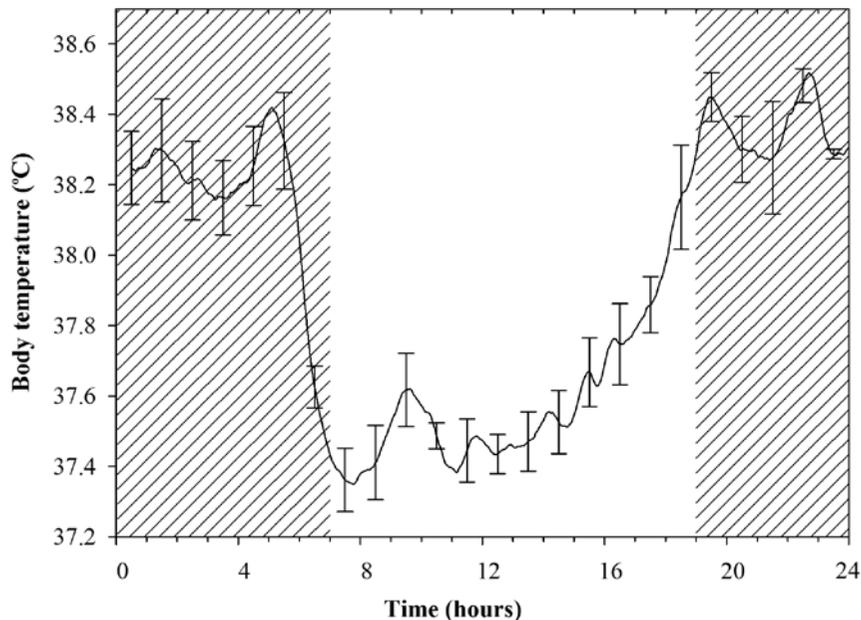


Fig. 5. A pattern of daily oscillation of body temperature in the Wistar albino rat (mean \pm SE of 5 animals, each averaged over 6 consecutive days). Data set of each animal were smoothed by a 30 min moving-averages filter to eliminate high-frequency oscillations and the average of 5 animals then computed. Data resolution is 1 min, although SE's are shown at hourly intervals to facilitate visual inspection. Lights were on daily from 7:00 to 19:00 hr.

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