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LETTER TO THE EDITOR

**Comparisons Between Periodograms and Spectral Analysis:
Don't Expect Apples to Taste Like Oranges**

This article on the analysis of time-series data attempts briefly to clarify certain important issues that were neglected by Dowse & Ringo (1989), in a paper entitled, "The Search for Hidden Periodicities in Biological Time Series Revisited." The central theme of their paper is that in general, additive methods of data analysis (all related to the classical periodogram† described by Schuster, 1898) are inferior tools for the analysis of biological-rhythm data, a conclusion with which I take issue.

The choice among methods for analysis was made by Dowse & Ringo (1987, 1989; see, also, Dowse *et al.*, 1987) on a simple and straightforward basis: MESA (Maximum Entropy Spectral Analysis), which they have championed, supports their claim that the *per^c* strain of *Drosophila melanogaster* shows endogenous ultradian rhythms (periods of a few hours); classical periodogram analysis *s.s.* does not confirm that interpretation, which is taken to be a "serious error of omission"; MESA has thus been shown to be a superior analytical tool, and "dependence on the Whittaker-Robinson 'periodogram' should be abandoned." (Dowse & Ringo, 1989: 495). Unfortunately, the issue is not as clearcut as this prescription suggests; the two different methods of time-series analysis involve different calculational principles, they are based on somewhat different assumptions, and they are suitable for asking somewhat different questions. Because of those fundamental differences between the two sorts of analysis, it is not in the least surprising that they sometimes give different results; apples can't be expected to taste exactly like oranges. Rather than rejecting one method or the other, it is more fruitful to determine what those differences in results imply.

In order to appreciate the complex issues involved, consider examples of two fundamentally different sorts of periodic processes: oceanic tides; and the waves generated by tossing pebbles into a pond at irregular intervals. Although the time scales are very different, both kinds of data-generating process would lead to plots of water height against time, in which repetitive oscillations would be evident; both processes produce periodic output. For purposes of analysis, the most important

†Dowse & Ringo (1989) refer to a plot of $f(T)$ vs. T from Fourier analysis (where T is period) as "the true periodogram", and when referring to the output of additive analysis, they place quotation marks around "periodogram." In my opinion, this usage can only lead to confusion. As demonstrated here, it is both useful and important to distinguish between additive and multiplicative methods of time-series analysis; and because the term "periodogram" has a 90-year history of application to the results of the additive method pioneered by Schuster (1898), I urge that some other term such as "spectrum" be used for plots of the results from Fourier analysis and related multiplicative methods. In this article, the abbreviation *s.s.*, for *sensu stricto*, will be appended (periodogram *s.s.*) to emphasize that terminological priority.

of the several expected differences between the two time-series records involves phase. In the pebble-generated waves, phase is not long conserved; the next pebble may strike at any time, and so its resulting waves are apt to be out of register with the preceding set. In contrast, because of the nature of the planetary forces driving the tides, phase of the tidal components is rigorously conserved, and tides are therefore predictable years in advance.

This difference in the nature of the data-generating processes has a profound influence on the usefulness of alternative methods of data analysis and their interpretation. Periodogram analysis *s.s.* is a linear (additive) procedure, and was designed to deal with situations in which phase is conserved (tides); alternative kinds of procedures that can collectively be designated as spectral analysis are non-linear (multiplicative) methods and are particularly useful for cases in which phase is not conserved. In periodogram analysis *s.s.*, a given measurement in the series is added to selected subsequent values (e.g. hourly averaging); in non-linear methods, a given measurement is multiplied by selected subsequent values (autocorrelation function). Each of these two basic kinds of analysis has its strengths and weaknesses.

The additive techniques have a long and successful history (Enright, 1981). The basic principle and its rationale were described and demonstrated by Schuster (1898); and thorough mathematical treatments of periodogram analysis *s.s.* were provided by Whittaker & Robinson (1924) and Kendall (1946). This sort of analysis has subsequently been elaborated to permit estimation of "energy" for those period values that are non-integer multiples of the basic time unit of the measurement series; and it has been extensively tested with a variety of artificial data inputs and with real biological-rhythm data (Enright, 1965*a, b*; Binkley *et al.*, 1973; Binkley, 1976; Rawson & DeCoursey, 1976). The periodogram *s.s.* has a number of weaknesses, which have been examined in detail (Enright, 1965*a*) and it should—like any statistical procedure—be used with caution, and with careful screening for possible artifacts and sources of misinterpretation.

Direct-inspection of data from the most common sorts of circadian rhythms (actograms) leaves no doubt that in these kinds of data, phase typically is conserved. The data-generating process leads to something approaching a stable spectral line, with no spontaneous phase "jumps". The output is more comparable with a record of the tides than with the waves resulting from pebbles thrown into a pond at random intervals. Hence, periodogram analysis *s.s.* is entirely appropriate for such data. Furthermore, given that a strong circadian rhythm is present in a data set, periodogram analysis *s.s.* can, even with relatively short records, give estimates of period that agree remarkably well (to 3 or 4 significant figures) with estimates derived from zero-crossing analysis (least-squares regressions) (Enright, 1965*a*: 444-445); similarly good agreement with expectation (to 3 or 4 significant figures) was also obtained in simulations with known input and superimposed additive noise (Enright, 1965*a*: 434-435). Multiplicative methods of time-series analysis do not usually provide this kind of resolution from short records. Hence, the unsupported assertion of Dowse & Ringo (1989: 495) that periodogram analysis *s.s.* "is low in resolution", compared with "more sophisticated techniques" is unwarranted. Instead, the evidence available (Enright, 1965*a*, 1981; Binkley *et al.*, 1973; Binkley, 1976)

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indicates that for a stable, phase-conservative rhythm, the accuracy and precision of period estimates obtainable from periodogram analysis *s.s.* are unsurpassed. Many different statistical methods can demonstrate rigorously that a given time-series is non-random; and other tests can be applied to determine whether there is statistically significant "energy" present in a given part of the spectrum. It is important to note, however, that no sort of time-series analysis can provide rigorous means of *proving* that a "rhythm" with a particular period is present in the data. That is, instead, an interpretational issue for which some arbitrary definition of "rhythm" is required. I chose periodogram *s.s.* procedures (actually, I unwittingly re-invented and elaborated on Schuster's method: Enright, 1965a) for a re-analysis of certain sets of data from the laboratories of Professor Frank A. Brown, Jr., for two reasons: because the calculational procedure represents nothing more than a generalization of the method of data analysis that had been used by the original authors to support their claims that precise, externally driven 24-hr and 24.8-hr rhythms existed in the data; and because even with relatively short data sets, the method provides good resolution, permitting a relatively precise estimate of the period of a persistent, phase-conservative rhythm, should one exist. Beyond this, the procedure is simple to understand at an intuitive level. The hypothesis under investigation in those re-analyses—that the sun and the moon provide the driving forces behind accurate biological rhythms presumably demonstrated under "constant" laboratory conditions—clearly implies conservation of phase: a situation comparable with oceanic tides, and not with pebbles thrown at random into a pond. I stand by my opinion that periodogram analysis *s.s.* provided the most appropriate test of that hypothesis.

Dowse & Ringo (1989) have demonstrated that when a multiplicative analysis is applied to those same data sets, the resulting spectra in some cases look quite different from the periodograms *s.s.*, and lend themselves to slightly different interpretations. Some of those differences may well reflect oscillatory components in which phase is not conserved—irrelevant for the "exogenously-driven" hypothesis being tested; others, I think, reflect problems inherent in the Dowse-Ringo procedures. For example, periodogram analysis *s.s.* had indicated that certain fiddler-crab data involved oscillations with period length appreciably greater than that of the tides (Enright, 1965a: figs 18b, 18c and 18e) but Dowse & Ringo (1989: 501-502) emphasize that MESA "gives the true tidal period of 12.4 hr in all cases." The conceptual difference between a *circa-tidal* rhythm and an *exact tidal* rhythm under laboratory conditions was the central issue of concern in my re-analyses of Brown's data (Enright, 1965a). From the additive analysis, the periods estimated for the rhythms were in several cases clearly different from that of the tides; from MESA, the estimated periods were indistinguishable from that of the tides (Dowse & Ringo, 1989). Which interpretation is correct? Simple graphs based on the raw data (Enright, 1965a: fig. 19) support the interpretation from periodogram analysis *s.s.*: when peaks in a "tidal rhythm" gradually change their timing by as much as 9 hr relative to the postulated driving force over an interval of 2 to 4 weeks, the interpretation of endogenous, free-running rhythmicity seems preferable to the alternative that the rhythmicity has a true period exactly that of the tides and may therefore be directly

evoked by lunar-tidal stimuli. In this case, the greater resolution of the periodogram *s.s.* seems clear; it confirms a phenomenon that can be demonstrated in the raw data, and that is not evident from MESA treatment.

Despite the widespread usefulness of periodogram procedures *s.s.* for the analysis of biological-rhythm data, these methods have one telling weakness: they are totally inappropriate for data in which phase is not conserved. The wave record from pebbles thrown into a pond, if treated by additive analysis, will provide no useful information at all about oscillatory properties of the system, unless segments of the record can be synchronized, prior to analysis, on the basis of the time that each pebble strikes the surface. (In some cases, such "artificial synchronization", followed by additive analysis, has proven to be a very useful procedure, as in post-stimulus averaging of sensory-receptor output or post-stimulus analysis of brain waves.) For many situations in which time-series analysis is contemplated, ranging from wind-generated waves on the ocean to an ongoing electroencephalogram, phase of the various components is not long conserved, and imposed synchronization of segments of the record is not possible; and is for such cases that non-linear methods of frequency analysis are essential. Modern rigorous understanding of non-linear methods was greatly advanced by Blackman & Tukey (1959); and subsequent developments have led to many variations in the techniques available. The Maximum Entropy Spectral Analysis advocated by Dowse & Ringo (1989) is such a procedure.

Central to all those techniques is the concept of Fourier analysis: the mathematical fact that almost any finite series of regularly spaced values can be represented by the sum of a series of sine and cosine functions with frequencies that are integer multiples of $1/N$, where N is the number of data points available. Although the various multiplicative procedures for time-series analysis based on Fourier analysis have proven to be broadly useful tools, a Fourier analysis is not the only valid way of summarizing a data sequence; an equally valid and general alternative would be a polynomial with N terms. Nor is Fourier analysis necessarily the simplest or most appropriate way of describing an oscillatory time-series. Suppose that the data-generating process represents an on-off switch, with 50% duty cycle, activated at regular intervals. The resulting square-wave output *can* be represented as the sum of a large number of (sinusoidal) Fourier components, based on the principal frequency and its odd harmonics; this is a mathematically valid analysis, but it would obscure rather than reveal the nature of the data-generating process. (Periodogram analysis *s.s.* makes no assumption about the wave-form of the data-generating process, and would, in this kind of case, directly demonstrate a single on-off wave-form.)

Spectral-analysis, based Fourier components, is not restricted to data sets in which phase is not conserved; it can also be applied to phase-conservative oscillations like ordinary circadian rhythms. One of the major disadvantages of applying such methods to biological-rhythm data, however, is that the fundamental estimates of frequency in the region of interest are quite coarse. Suppose that one has a 10-day record of hourly values: then the first few "components" into which the record is resolved have periods of 240, 120, 80...26.67, 24, 21.82 hr, etc (i.e. $N/1, N/2, N/3, \dots$); and some elaboration of the basic method is necessary in

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order to obtain a more refined estimate of the period of the data-generating system that is responsible for the spectral energy assigned to any given band. With a spectral peak at, say, 24 hr one knows only that the best estimate of period is somewhere within the range between 21.8 and 26.7 hr. In the typical application of multiplicative spectral analysis by engineers or physicists to data from non-biological sources, this limitation on precision does not represent a serious restriction: one simply extends measurement over enough cycles that this $1/N$ limit on resolution is no problem; but that solution is usually not convenient in circadian-rhythm studies. MESA is one of several refinements of multiplicative time-series analysis that attempt to circumvent the theoretical limit on frequency resolution that is inherent in Fourier analysis; one obtains finer resolution in the estimates of period, but sacrifices meaningful confidence limits for the estimates, such as those associated with the Blackman-Tukey power spectrum (Blackman & Tukey, 1959). On the other hand, periodogram analysis s.s. makes no pretense of providing confidence limits for estimates of period.

Dowse and Ringo (1989: 495) advocate general abandonment of periodogram procedures s.s. because MESA treatment of certain *Drosophila* data indicates the presence of ultradian rhythmicity (periods between 4 and 22 hr), and periodogram analysis s.s. did not, in general, support that interpretation. Instead of rejecting either method of analysis, it is more interesting to inquire what this difference in outcomes may imply. In view of the differences in principle between additive and multiplicative methods of analysis, a plausible interpretation would be that the insects may show short-period "oscillations" in which phase is not conserved: comparable with those from sets of waves caused by pebbles thrown into a pond at random times or with the voltage waves of an electroencephalogram. If the flies studied by Dowse *et al.* (1987) do indeed show ultradian periodicity with non-conservative phase, the question of whether such behavior deserves to be called a "rhythm" and to be compared with circadian rhythmicity is an interesting interpretational issue that goes beyond the scope of time-series analysis. In any case, it is not evident to me that the re-re-analyses by Dowse & Ringo (1989) of the data treated in Enright (1965a) have led to significant new interpretations of the data involved, or contributed new insights about the now largely forgotten endogenous-exogenous controversy that prompted my treatment.

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