

1988, Stewart and Zeigler 1984, Zeigler and Stewart 1977). Both *I. frontalis* and *I. zionensis* duets have been described by these authors as derived, complex symphonic conversations. The calls of *I. elongatus* described by Stewart and Zeigler (1984) appeared to be monophasic ancestral calls, but were based on only six signals from one male, therefore the authors stated that they could only be considered a preliminary characterization of the species. The objective of this study was to rear virgin males and females of all *Isogenoides* species, determine their call and answer signals and duet patterns, and examine comparatively these patterns within the genus and how they fit with the evolutionary paradigm of Stewart (2001).

METHODS AND MATERIALS

Virgin adults of eight species were reared from slightly mature to mature larvae and collected as follows: (1) *Isogenoides colubrinus* (Hagen), Yampa River, At Twelvemile Gulch and Cross Canyon National Park Service area, Moffat Co., Colorado, 13/III/2002; (2) *I. doratus* (Frison), Pine River, At Walker's Bridge, 2 mi NE. of Skookum on State Rd., Lake Co., Michigan, 11/IV/1999; (3) *I. doratus*, Rock River, At Lakewood Corner, bridge on Hwy. 75, 7.5 mi. S. of Rock Rapids, Lyon Co., Iowa, 09/II/2002; (4) *I. elongatus* (Hagen), Colorado River, Bridge on Hwy 40, 2.5 mi. W. of Granby, Garfield Co., Colorado, 01/VI/1999; (5) *I. elongatus*, Colorado River, At Rifle City Park, Garfield Co., Colorado, 12/III/2000; (6) *I. frontalis* (Newman), Rocky Run Creek, At Cty. Hwy. H, 1.5 mi. N. of Brule, Douglas Co., Wisconsin, 07/IV/2000; (7) *I. frontalis*, Cranberry River, Near intersection of Carmichael and Cranberry Rds., 3 mi. S. of Herbster, Bayfield Co., Wisconsin, 08/IV/2000; (8) *I. frontalis*, Pine Creek, At confluence of Fish Creek at Fish Cr. Rd. and Old US 2, Bayfield Co., Wisconsin, 09/IV/2000; (9) *I. hansonii* (Ricker), Stony Creek, At CR 635, 2 mi. N. of Laurel Branch confluence, Jefferson N.F., Giles Co., Virginia, 27/II/2001; (10) *I. olivaceus* (Walker), Pere Marquette River, Bridge on Hwy 37, 2.5 mi. S. of Baldwin, Lake Co., Michigan, 10/IV/1999; (11) *I. olivaceus*, Pine River, At Walker's Bridge, 2 mi NE. of Skookum on State Rd., Lake Co., Michigan, 11/IV/1999; (12) *I. varians* (Walsh), Westville Creek, Bridge on Hwy 43 at Pinola, Simpson Co., Mississippi, 12/II/2000; (13) *I. varians*, Big Otter River, Bridge on Hwy. 24 near intersection of CR 709 and CR 711, 2 mi. W. of Cambell Co., line, Bedford Co., Virginia, 26/II/2001; (14) *I. zionensis* Hanson, San Miguel River, Near Placerville, San Miguel Co., Colorado, 13/VI/1999.

Drumming signals were recorded with a Marantz portable cassette recorder (model PMD430) and Sony electret condenser microphones (model ECM95S) or with a Sony MiniDisc portable recorder (models MZ-R37 or MZ-R700) and Optimus omnidirectional microphones (model 33-3013), in a 2-compartment, glass covered field recording chamber (Stewart and Sandberg In Press, Sandberg and Stewart 2003) Recorded signals were encoded and analyzed using the methods provided in (Sandberg and Stewart 2003).

RESULTS

Isogenoides colubrinus

One hundred fifteen and 114 signals were obtained from five 1-6 day old males and four females, respectively, at 20-28°C, or normal incandescent or fluorescent room lights (Provo, Utah and Brighton, Colorado), and partially shaded sunlight near mountain streams (Leopard Creek and Mosquito Creek, Colorado). Their exchanges were complex, ranging from 3-way call-answer-response sequences (N=12) (Fig. 1A), to grouped calls with grouped, interspersed female answers (Fig. 1B). Typically, the grouped exchanges involved a short symphony of about three M-F volleys, terminated by a slow (wider-interval) male response signal. Females did not always intersperse an answer between every male call group and two exchanges lacked the terminal response group.

The male sequenced and grouped calls consisted of five mode beats (5.2 ± 0.7) with intervals of 65.0 ± 8.3 msec (Table 1). These expressions [examples: (5.2 ± 0.7) and 65.0 ± 8.3 msec] represent a $\bar{x} \pm$ standard deviation and will be used as such throughout the rest of this paper. Only 10 symphonies possessed male call groups with seven beats, all others ranged from 3-6. The number of call groups ranged from 1-7, with individual mean interbeat intervals (interval 1 (i1) – interval 6 (i6)) generally increasing, earlier intervals increased slightly in approximate 3-msec increments from 59.7 ± 7.8 msec (1st) to 72.6 ± 9.0 msec (5th). The last mean interbeat interval decreased to 69.4 ± 5.7 msec, which may be suspect due to small sample size (N=10).

Mode and mean number of beats per interspersed female answer signals were 1 and 1.8 ± 0.8 msec (Table 1). Average interbeat intervals increased throughout the answer and overall they were 121.8 ± 26.8 msec. In duets and symphonies, female answers followed male call groups (M-F exchange interval) by 402.0 ± 101.2 msec and subsequent male calls followed female answer groups (F-M exchange interval) by 1654.8 ± 429.1 msec. The terminal male response signal was different from sequenced and grouped calls with a mode beat count of 2 (2.5 ± 0.9) and interbeat intervals of 441.8 ± 142.5 msec (Table 1). Mean intervals of the response decreased from 452.4 ± 136.2 msec (1st) to 384.0 ± 135.5 msec (4th), the last interval increased to 462.5 ± 147.8 msec, which may be suspect due to small sample size (N=2). The last (F-M) exchange interval was 626.4 ± 198.5 msec.

Isogenoides doratus

Two populations were recorded in 1999 and 2002, from Michigan and Iowa respectively. The Michigan tests included 126 exchanges obtained from one, 1-9 and 1-12 day old male and female respectively, at 24°C, and 88 FTC. Their exchanges were grouped male calls with interspersed female answers (Fig.2A). Typically, the exchanges involved long symphonies of about ten M-F grouped signals that ended either with an unanswered male call group or with a female answer signal. Males did not end exchanges with response signals and females did not always intersperse an answer between every male call.

The Michigan male call groups consisted of three mode beats (3.1 ± 0.6) with intervals of 16.7 ± 3.0 msec (Table 1). Only nine symphonies possessed male call groups with five beats, all others ranged from 1-4. The number of call groups ranged from 2-21 with individual mean interbeat intervals (i1 – i4) generally decreasing with irregular intervals from 17.5 ± 2.1 msec (1st) to 10.1 ± 1.8

msec (3rd). The last interval increased slightly, which may be suspect due to small sample size.

Mode and mean number of beats per interspersed female answer signals were 2 and 2.1 ± 1.0 (Tables 1 and 3). Average interbeat intervals increased throughout the answer and overall they were 107.0 ± 52.0 msec. In symphonies the (F-M) exchange interval was 699.4 ± 170.0 msec. Females began their grouped answer signals only after the second male call-group and the (M-F) exchange interval was 466.4 ± 89.4 msec.

The Iowa tests included 115 exchanges obtained from four and six, 2-7 day old males and females respectively, at 24°C, and 80-88 FTC. Their exchanges were grouped male calls with interspersed female answers (Fig. 2B) and similar to Michigan exchanges. Typically, the exchanges involved long symphonies of either about seven M-F signals that ended with an unanswered male call group or with a female answer signal. Males did not end exchanges with response signals and females did not always intersperse an answer between every male call.

The Iowa male call groups consisted of three mode beats (3.1 ± 1.2) with intervals of 14.4 ± 3.8 msec (Table 1). Only six and one symphonies possessed male call groups with seven and eight beats respectively, all others ranged from 1-6. This increased beat-resolution over the Michigan tests may be attributed to our then-new digital recording methods and minor changes made to the recording chamber. The number of call groups ranged from 2-22, with individual mean interbeat intervals (i1 – i7) decreasing unevenly from 16.7 ± 2.2 msec (1st) to 6.5 ± 1.3 msec (6th). The last interval increased, and may be suspect due to small sample size.

Mode and mean number of beats per interspersed Iowa-female answer signals were 1 and 1.5 ± 0.7 (Table 1). Average interbeat intervals decreased until the penultimate (4th) interval, then increased slightly during the 5th interval and overall they were 135.5 ± 40.7 msec, which is notably larger than Michigan average answer intervals. This unexplained decreasing interval pattern for Iowa females is opposite to the Michigan females. In symphonies the (F-M) exchange interval was 724.8 ± 169.2 msec. Females began their grouped answer signals after the second male call group, except one exchange, where the female answered the male's first call and the (M-F) exchange interval was 358.7 ± 79.9 msec. We propose, with the exceptions of overall average female interbeat interval, and opposite female answer interval patterns, that these two *I. doratus* populations compared well. Male mode and average number of beats and overall mean beat interval overlapped sufficiently.

Isogenoides elongatus

Two Colorado populations were recorded in 1999 and 2000 from two different sampling sites on the Colorado River, the first near Granby and the second at Rifle respectively. The Granby tests included 160 signals obtained from seven and three field collected males and females respectively, at 18-21°C, and normal incandescent room lighting in Pitkin, Colorado. The age of these individuals was unknown but assumed old because kick net sampling for nymphs was unsuccessful and no other adults were observed in usual habitats.

The Granby drumming exchanges were complex, ranging from 2- and 3-way call-answer-response sequences (Figs. 3A and 3B), to grouped calls with grouped, interspersed female answers (Fig. 3C). Typically, the sequences involved a single M-F volley that sometimes ended with a slower (1-2 beat) male response

signal. Females always interspersed an answer between every male call group and 111 duets lacked the slow male response signal.

The Granby sequenced and grouped male calls consisted of seven mode beats (6.8 ± 1.1) with intervals of 397.0 ± 75.0 msec (Table 1). Only six exchanges possessed male first call groups with 9-beats and one with 10-beats, all other male call groups ranged 3-8 beats. The number of call groups ranged from 1-3, with individual mean interbeat intervals ($i_1 - i_9$) increasing unevenly from 354.4 ± 51.8 msec (1^{st}) to 472.8 ± 88.5 msec (8^{th}). The last mean interbeat interval increased noticeably to 638.3 ± 44.6 msec, and may be suspect due to small sample size.

Mode and mean number of beats per sequenced and interspersed Granby female answer signals were 3 and 2.5 ± 0.6 msec (Table 1). Average interbeat increased throughout the answer and overall intervals were 386.9 ± 93.7 msec. In sequenced and grouped exchanges, the (M-F) exchange interval was 925.5 ± 286.0 msec. The terminal male response signals were different from calls with a mode beat count of 1 (1.1 ± 0.3) and mean interbeat intervals of 760.8 ± 222.7 msec (Table 1). The (F-M) exchange interval was relatively even throughout exchanges with an average of 1971.9 ± 159.8 msec. The last of these (F-M) exchange intervals, before the male response signal, decreased to 888.8 ± 294.8 msec.

The Rifle drumming exchanges were also complex, ranging from 2- and 3-way call-answer-response sequences to grouped calls with grouped, interspersed female answers (Fig. 3D). Typically, the sequences involved a single M-F volley that sometimes ended with a 1-beat male response signal (Table 1). Females did not always intersperse an answer between every male call group and 64 duets lacked the slow male response signal.

The Rifle sequenced and grouped male calls consisted of six mode beats (5.9 ± 0.7) with intervals of 222.1 ± 14.0 msec (Table 1). These call intervals were noticeably smaller than Granby call intervals and may be attributed to the assumed increased age of the Granby test pairs. Only one exchange possessed a first call group with 8-beats, all other male call groups ranged 4-7 beats. The number of call groups ranged from 1-3, with individual mean interbeat intervals ($i_1 - i_7$) increasing unevenly from 207.5 ± 17.9 msec (1^{st}) to 229.2 ± 9.0 msec (4^{th}). Interval 5 decreased slightly (228.0 ± 8.8 msec), then increased irregularly to the last interval (7^{th}) 242.1 msec ($N=1$). The interval pattern in general increased from the first to last beats for both populations. Incremental interval increase appeared to be greatest at the beginning and ending of average sequenced and grouped calls with less incremental increase in middle intervals.

Mode and mean number of beats per sequenced and interspersed Rifle female answer signals were 2 and 1.8 ± 0.5 msec (Table 1). Average interbeat intervals increased throughout the answer and overall they were 180.5 ± 20.7 msec. However, these intervals were shorter than the Granby population and assumed to be so because of their younger age. In sequenced and grouped exchanges, the (M-F) exchange interval was 570.0 ± 82.1 msec and the terminal male response signal was different from calls with a mode beat count of 1 (Table 1). The (F-M) exchange interval was relatively even throughout exchanges with an average of 950.2 ± 124.1 msec. The last (F-M) exchange interval decreased to 648.3 ± 178.6 msec.

In agreement with (Stewart 1997), male-call beat and interval data convey the critical information of species specificity, possible reproductive fitness, and

phylogenetic information. Therefore the two *I. elongatus* populations agree with one another in terms of the male call interval description. The major differences were the Granby males and females increased beat intervals that are assumed the result of their increased age.

Isogenoides frontalis

One hundred thirty-five exchanges were obtained from ten, 1-2 day old males and four females, respectively, at 23-24°C, and 84 FTC. Their exchanges were complex, ranging from 3-way call-answer-response sequences (Fig 4A) to grouped calls with grouped, interspersed female answers (Fig. 4B). Typically, the grouped exchanges involved a short symphony of about three M-F signals, terminated by a slow male response signal. Females did not always intersperse an answer between every male call group and three duets lacked response signals.

The male sequenced and grouped calls consisted of four mode beats (4.1 ± 0.7) with intervals of 61.2 ± 11.3 msec (Table 1). Only one symphonic exchange possessed a male group with six beats, all other male call groups ranged 2-5 beats. The number of call groups ranged from 1-6, with individual mean interbeat intervals (i1 – i5) generally increasing at approximate 8-msec increments, from 51.9 ± 8.4 msec (1st) to 77.3 ± 8.3 msec (4th). The last interval decreased to 65.5 msec (N=1) that may be considered suspect due to small sample size.

Mode and mean number of beats per interspersed female answer signals were 2 and 1.6 ± 0.6 msec (Table 1). Average interbeat intervals decreased throughout the short answers and overall they were 93.8 ± 29.3 msec. In symphonic exchanges, the (M-F) exchange interval was 581.5 ± 76.3 msec and the terminal male response signal was different from call groups with a mode beat count of 3 (3.5 ± 1.5) and interbeat intervals of 288.8 ± 129.4 msec. (Table 1). Mean intervals of the response increased from 251.8 ± 114.3 (1st) to 446.2 msec (N=1) (7th), the sixth interval decreased to 297.0 ± 38.7 msec and these last two intervals may be suspect due to small sample size (N=4).

Isogenoides hansonii

Sixty signals were obtained from four, 2-5 day old males at 22-24°C, and 70 FTC. Their calls were grouped signals with rapid interbeat intervals and short resting intervals between groups (Fig. 5). Typically, the first group contained more beats than the last two, with greatest interval variation within the first five beats. Female nymphs collected at the same time as males (February) failed to emerge successfully (April) under prolonged simulated-stream laboratory conditions.

The male grouped calls consisted of six mode beats (9.3 ± 4.3) with intervals of 31.3 ± 9.4 msec (Table 1). Only four calls possessed groups with beat counts of 19, all other ranged from 5-18. All male calls contained three groups, with individual average interbeat intervals (i1 – i18) decreasing from 35.0 ± 13.8 msec (1st) to 29.0 ± 5.4 msec (6th), then remaining approximately even over intervals 7-11 with irregular increases and decreases of 0.2-0.9 msec. A decreasing interval pattern was again observed from the (12th) 28.8 ± 2.1 msec to the (17th) 25.0 ± 4.0 msec (N=11). The last interval increased slightly to 25.6 ± 1.2 msec, which may be suspect due to small sample size. The intervals between male calls (...B-...B exchange interval) remained approximately even at 259.2 ± 21.1 msec and average beat counts decreased, the first group with 14.2 ± 2.7 , the second with $7.3 \pm$

0.7, and the last with 5.8 ± 0.6 . The average interbeat intervals for each group overall decreased from 33.0 ± 12.3 msec (1st) to 28.8 ± 2.1 msec (2nd), and then increased slightly to 29.9 ± 2.1 msec (3rd).

Isogenoides olivaceus

Two hundred sixty-nine signals were obtained from two 2-12 day old males and seven females, respectively, at 20-24°C, and 74 FTC. Their exchanges were mostly simple, ranging from 2-way (Fig. 6A) to 3-way call-answer-response sequences (Fig. 6B). Only one grouped 7-way exchange (Fig. 6C) was recorded over an eleven-day drumming period. Typically, the sequences involved a single M-F volley that sometimes (N=88) ended with a slower male response signal.

The male sequenced calls consisted of seven mode beats (6.7 ± 0.5) with intervals of 98.2 ± 12.0 msec (Table 1). Only three male-calls possessed call groups with 8-beats, all others ranged from 5-7. The number of call groups ranged from 1 (N=269) to 3 (N=1), with individual sequenced mean interbeat call intervals (i1 – i7) decreasing irregularly from 107.0 ± 12.0 ms (1st) to 85.8 ± 8.2 ms (7th). Average incremental interval decrease was slow during the first three intervals, then rapid during intervals 4 and 5, and finally slow again over the remaining intervals.

Mode and mean number of beats per sequenced female answer signals were 6 and 5.5 ± 1.7 msec (Table 1). Average interbeat intervals increased from 74.9 ± 16.4 msec (1st) to 115.1 ± 32.9 msec (7th), decreased slightly at the penultimate (8th) interval (113.5 ± 37.2 msec), and then increased to 126.0 ± 66.7 msec (9th), and overall they were 86.6 ± 22.0 msec. In sequences, the (M-F) exchange intervals were 739.9 ± 130.9 msec and (F-M) exchange intervals were 831.6 ± 231.1 msec. The terminal male response signal was different from calls with a mode beat count of 1 (1.9 ± 1.0) and interbeat intervals increased from 340.2 ± 143.9 msec (1st) to 517.0 (N=1) (5th).

Isogenoides varians

Two populations were recorded in 2000 and 2001, from Mississippi and Virginia respectively. The Mississippi tests included 125 signals from four 4-10 day old males and four females respectively, at 20-24°C, and 74 FTC. One recorded female did not answer the calls of two different males. Their exchanges were complex, ranging from 3-way call-answer-response sequences (N=3), to grouped calls with grouped, interspersed female answers (Fig. 7A). Typically, the grouped exchanges involved a short symphony of about three M-F volleys, terminated by a slow male response signal. Females did not always intersperse an answer between every male call and only answered the first-male call group in four exchanges. Males did not always end sequenced or grouped exchanges with the slow response group.

The Mississippi male sequenced and grouped calls consisted of six mode beats (6.5 ± 1.6) with intervals of 26.7 ± 3.1 msec (Table 1). Only four symphonies possessed male calls with 10 beats, all others ranged from 2-9. The number of male sequenced and grouped calls ranged from 1-7, with individual mean interbeat intervals (i1 – i9) approximately even with only slight interval changes from 25.8 ± 3.7 msec (1st) to 28.0 ± 2.9 msec (6th and 7th), decreasing slightly to 27.1 ± 2.2 msec (8th), then finally increasing again to 30.0 ± 1.6 msec (9th).

Mode and mean number of beats per interspersed Mississippi female were 4 and 3.4 ± 1.0 (Table 1). Average interbeat intervals increased throughout the

answer and overall they were 52.4 ± 9.2 msec. The (M-F) exchange interval was 944.8 ± 64.0 msec and the (F-M) exchange interval was 1285.7 ± 276.3 msec. The terminal male response signal was different from sequenced and grouped calls with a mode beat count of 3 (3.2 ± 1.3) and interbeat intervals of 56.0 ± 11.8 msec (Table 1). Mean male response intervals increased from 53.5 ± 11.7 msec (1st) to 59.3 ± 11.1 msec (4th), and then decreased to 46.6 ± 3.6 msec and 47.7 msec (5th and 6th respectively) that may be suspect due to small sample size (N=3).

The Virginia tests included 117 signals obtained from four 1-12 day old males and two females respectively, at 24°C, and 76 FTC. Their exchanges were complex and ranged from 2-way (N=2) and 3-way (N=4) call-answer-response sequences to grouped calls with grouped, interspersed female answers (Fig. 7B). Typically, the grouped exchanges involved a short symphony of about three M-F volleys, terminated by a slow male response signal. Females did not always intersperse an answer between every male call and only answered the first-male call group in seventeen exchanges. Males did not always end sequenced or grouped exchanges with the slow response group.

The Virginia male sequenced and grouped calls consisted of eight mode beats (7.1 ± 1.5) with intervals of 36.9 ± 4.0 msec (Table 1). Only five symphonies possessed male call groups with 10 beats, and two with 11 beats, all others ranged from 2-9. The number of male sequenced and grouped calls ranged from 1-6, with individual mean interbeat intervals (i1 – i10) slightly increasing with small interval changes from 35.4 ± 4.5 msec (1st) to 41.7 ± 5.0 msec (8th), decreasing to 40.2 ± 5.3 msec (9th), then finally increasing to 44.4 ± 2.2 msec (10th). The last three call-intervals may be suspect due to smaller sample size.

Mode and mean number of beats per interspersed Virginia female were 2 and 2.4 ± 0.9 (Table 1). Average interbeat intervals increased from 52.6 ± 7.1 msec (1st) to 70.4 ± 11.0 msec (3rd), and then decreased to 58.6 msec (N=1). This last interval may be suspect due to small sample size. The (M-F) exchange interval was 839.9 ± 61.4 msec and the (F-M) exchange interval was 1279.4 ± 305.2 msec. The terminal male response signal was different from sequenced and grouped calls with a mode beat count of 3 (3.1 ± 0.9) and interbeat intervals of 73.4 ± 21.4 msec (Table 1). Mean male response intervals increased from 63.7 ± 11.1 msec (1st) to 94.2 ± 35.8 msec (3rd), and then decreased to 71.8 msec (N=1). This last interval is suspect due to small sample size.

The differences in mode male number of beats and overall male call interbeat interval are substantial and suggest that these

I. varians populations may have unique drumming dialects. Differences could also be the result of unknown local environmental variables, such as temperature, atmospheric pressure, or light intensity. Another possibility for difference is that these two populations may be changing and one may become a potentially behaviorally cryptic species (Sandberg and Stewart 2003).

Isozenoides zionensis

Three hundred and seven signals were obtained from seven 2-5 day old males and nine females respectively, at 21-22°C, and normal room lighting in Pitkin, Colorado. Their exchanges were complex and ranged from 2- or 3-way call-answer-response sequences (Fig. 8A), to grouped calls with grouped, interspersed female answers (Fig. 8B). A third and more complex drumming exchange type was observed (N=21) where males linked together two or more simple call-answer-response sequences by beginning their next call series within or soon after

the otherwise terminal response signal (Fig. 8C). Usually (less complex sequenced and grouped exchanges), after sending the terminal response sequence, males began moving (localized search behavior) for variable lengths of time, while females remained stationary, and then after searching, continued with new call signal. Several exchanges containing these (male response-male call) variable series show a decreasing trend, shortening the time lag between the male response and next call. Therefore, these complex M-F exchanges contained series of grouped or sequenced, or both grouped and sequenced exchange types. Typically, grouped exchanges involved a short symphony of about four M-F volleys, terminated by a slow male response signal. Females did not always intersperse an answer between every male call group and males did not always end sequenced or grouped exchanges with the slow response group.

In complex series where male response signals were linked to the following male call signals, the average M(call) - M(response) exchange intervals were 458.3 ± 248.7 msec. These intervals were shorter than the M(call) - M(call) exchange intervals between grouped and unanswered male call groups and were 1083.5 ± 266.8 msec.

The less-complex male sequenced and grouped calls consisted of four mode beats (4.0 ± 0.6) with intervals of 364.3 ± 59.1 msec (Table 1, first entry). Only one sequence possessed a male call group with six beats, all others ranged from 1-5. The number of call groups ranged from 1-3, with individual mean interbeat intervals ($i_1 - i_5$) generally switching from increasing to decreasing. Intervals increased from 337.6 ± 71.0 msec (1^{st}) to 380.3 ± 46.8 msec (3^{rd}), and then decreased noticeably to 359.8 ± 25.8 msec (4^{th}). The final interval decreased to 344.4 msec, which may be suspect due to small sample size. However, in terms of both sample size and amount of change, we interpret this as the first switching interval pattern observed in *Isogenoides* male call groups. All other *Isogenoides* individual call intervals generally increased or decreased.

Mode and mean number of normal sequenced or interspersed female answer signals were 2 and 2.2 ± 1.1 msec (Table 1). Average interbeat intervals increased throughout the answer and overall they were 136.8 ± 32.7 msec. In symphonies, the (M-F) exchange intervals were 624.3 ± 129.3 msec and the (F-M) exchange intervals were 561.2 ± 197.3 msec. The terminal male response signals were different from sequenced and grouped calls with a mode and mean beat count of 3 (3.7 ± 1.4) and interbeat intervals of 132.2 ± 43.4 msec. Mean intervals of the response increased from 107.4 ± 24.6 (1^{st}) to 198.5 ± 56.9 msec (5^{th}).

Twenty one complex signals were obtained from two and five males and females respectively, under the same conditions as above. Their exchanges were the most complex observed in *Isogenoides* where the typical series was usually a grouped exchange type that involved a call-answer-response-call pattern as described above.

The more-complex male calls consisted of four mode beats (3.6 ± 1.1) with intervals of 344.8 ± 32.2 msec (Table 1, second entry). Only two series possessed call groups with six and seven beats respectively, all others ranged from 1-5. The number of call groups ranged from 2-6, with individual mean interbeat intervals ($i_1 - i_6$) generally switching from increasing to decreasing. Intervals increased from 327.2 ± 32.5 msec (1^{st}) to 356.5 ± 26.4 msec (3^{rd}), and then decreased from 347.3 ± 28.9 msec (4^{th}) to 329.0 msec (5^{th}). The final interval increased slightly to 335.0 msec and these last two intervals are suspect due to small sample size.

Mode and mean number of beats per interspersed complex-female answer signals were 2 and 1.9 ± 0.9 msec (Table 1, second entry). Average interbeat intervals increased throughout the answer and overall they were 123.4 ± 75.3 msec. In symphonies, the (M-F) exchange intervals were 623.1 ± 78.6 msec and the (F-M) exchange intervals were 508.4 ± 166.7 msec. The terminal male response signals were different from sequenced and grouped calls with a mode and average beat count of 4 (3.8 ± 1.1) and interbeat intervals of 135.3 ± 78.2 msec. Mean intervals of the response increased from 96.3 ± 16.7 (1st) to 299.3 ± 26.4 msec (5th). These data for complex-exchanges are compatible with all previous descriptions of this complex form of drumming. Only the elucidation of a switching interval description and M(call) - M(response) pattern are new information for this species.

One *I. zionensis* female answered male calls with a signal type other than tapping her abdomen. She fluttered her wings and her abdomen vibrated against the substrate for periods ranging from 2035.0 – 5352.9 msec. This wing-flutter behavior occurred after the male called from 1 to three times and often overlapped the male's next call.

Two recording sessions of this species took place during the morning when room temperature remained at 66° F (18.9° C). The two experimental pairs appeared to drum normally until analysis indicated a substantial difference in overall average call interval. One hundred thirty-eight signals were obtained from two 3-4 day old males and two females respectively, under normal room lighting in Pitkin, Colorado. Their signal types ranged from 3-way sequences to long grouped exchanges, some as long as 5-way exchanges.

The male sequenced calls and grouped exchanges consisted of four mode beats (4.2 ± 0.5) with overall average intervals of 589.2 ± 50.7 msec (Table 4). The number of call groups ranged from 1-3, with individual mean interbeat call intervals (i1 – i4) increasing irregularly.

Mode and mean number of beats per female answer signals were 2 and 2.3 ± 1.1 msec (Table 2). Overall average interbeat intervals were 206.7 ± 54.9 msec and the male response intervals averaged 184.7 ± 37.9 msec.

Isogenoides zionensis male and female sequenced and grouped exchanges at two temperatures (21–22°C and 18.9°C) had similar mode number of call- and answer-beats (Table 1 and Table 2), but overall beat intervals were larger for calls, answers and responses of colder males and females (Table 1 and Table 2), as would be predicted for poikilotherms.

DISCUSSION

The overall summary of signal characters (Tables 1, 2) shows that when all parameters of male calls, female answers and duetting characteristics are considered, that *Isogenoides* drumming is species-specific. There is some overlap of beat intervals in two pairs: (1). *I. colubrinus* and *I. frontalis*, (2). *I. hansonii* and *I. varians*. This might be expected in the first pair because of their morphological closeness, the fact that they were previously considered a single species (Hanson 1943) or subspecies (Ricker 1952), and that their ranges have little overlap (except in Saskatchewan). The largely allopatric populations possibly have not been under selective pressure to diverge the beat interval parameter of their signals. The two *I. varians* populations appeared different in average beat intervals, pos-

sibly explained as geographically determined dialects. Although the average beat intervals of *I. hansonii* and *I. varians* show some overlap, there may be specificity in the actual change of intervals during calls. Those of *I. hansonii* decrease, while those of *I. varians* show slight increase. Additionally, when comparing the average call beat-count per group, *I. hansonii* is slightly larger (9.3 ± 4.3) than *I. varians* (6.5 ± 1.6). Further testing is necessary in order to confirm that beat-counts and actual intervals contain species-specific information recognized by these ♀ females.

Isogenoides drumming signals ranged from sequenced 2- to 3-way M(call) - F(answer) or M(call) - F(answer) - M(response) exchanges to grouped male calls with grouped, interspersed female answers, terminated with or without a male response. One species, *I. zionensis* also had complex exchanges that may be described as containing long series of grouped or sequenced, or both grouped and sequenced exchange types. Pair-forming drumming characteristics and polarity of male-female exchange patterns (sequenced or non-multigrouped vs. grouped or multigrouped) were analyzed among six other drumming characters using an out group comparison (Watrous and Wheeler 1981) by Maketon and Stewart (1988). They determined that beat grouping was a strong derived character and that sequenced exchanges were ancestral by application of the "commonality principal" among all Arctoperlarian families (In-group "common = primitive") (Watrous and Wheeler 1981).

Therefore, drumming in *Isogenoides* can generally be characterized as derived to varying degrees in all species, except *I. olivaceus*, since their calls, male-female exchange patterns and female answers contain at least some signal grouping. *Isogenoides olivaceus* exchanges were almost entirely ancestrally sequenced (N=269) with only one grouped exchange recorded in an eleven-day drumming period.

The calls, male-female exchanges and answers of four species: *I. colubrinus*, *I. elongatus*, *I. frontalis*, and *I. varians*, contained both sequenced (ancestral) and grouped (derived) elements, and therefore appear to represent an intermediate transition in evolution of signaling (Stewart 2001) within the genus. The calls of *I. hansonii* (females were not successfully recorded) and calls, male-female exchanges and answers of both populations of *I. doratus* were exclusively grouped, and may therefore be considered derived and further specialized. The complex *I. zionensis* symphonies represent a third pattern of specialization, having both the sequenced ancestral and grouped elements for both sexes, and addition of two unique characters: 1. grouped portion containing a switching interval pattern, and 2. the inclusion of a male terminal response signal immediately preceding a continuing call group (Fig. 8). The drumming of all *Isogenoides* species fits at some level of the Stewart (2001) evolutionary paradigm, and the discussion of evolution of drumming in Perlodidae by Maketon and Stewart (1988).

LITERATURE CITED

- Bailey, W.J. 2003. Insect duets: underlying mechanisms and their evolution. *Physiol. Entomol.* 28: 157–174.
- Hanson, J.F. 1943. Records and descriptions of North American Plecoptera. Part III. Notes on *Isozenoides*. *Amer. Midl. Natur.* 29: 657–669.
- Maketon, M. and K.W. Stewart. 1988. Patterns and evolution of drumming behavior in the stonefly families Perlidae and Peltoperlidae. *Aquatic Insects* 10: 77–98.
- Maketon, M., K.W. Stewart, B.C. Kondratieff and R.F. Kirchner. 1988. New descriptions of drumming and evolution of the behavior in North American Perlodidae (Plecoptera). *J. Kans. Entomol. Soc.* 61: 161–168.
- Ricker, W.E. 1952. Systematic studies in Plecoptera. *Indiana Univer. Publ. Sci. Ser.* 18: 105–114.
- Sandberg, J.B. and K.W. Stewart. 2003. Continued studies of drumming in North American Plecoptera; Evolutionary implications, pp. 73–81. *In* E. Gaino (ed.), *Research update on Ephemeroptera and Plecoptera*. University of Perugia, Perugia, Italy.
- Sandberg, J.B. and K.W. Stewart. 2004. Description of the nymph and drumming calls of *Megaleuctra complicata* Claassen (Plecoptera: Leuctridae); Evolution of drumming in Leuctridae. *Aquatic Insects* 26: 123–129.
- Stewart, K.W. 1994. Theoretical considerations of mate finding and other adult behaviors of Plecoptera. *Aquatic Insects* 16: 95–104.
- Stewart, K.W. 1997. Vibrational Communication in Insects: Epitome in the Language of Stoneflies? *Amer. Entomol.* 43: 81–91.
- Stewart, K.W. 2001. Vibrational communication (drumming) and mate-searching behavior of stoneflies; Evolutionary considerations. pp. 217–226. *In*: *Trends in research in Ephemeroptera and Plecoptera*. Dominguez, E., (ed.) Kluwer Academic/Plenum Publishers, NY.
- Stewart, K.W. and M. Maketon. 1991. Structures used by Nearctic stoneflies (Plecoptera) for drumming, and their relationship to behavioral pattern diversity. *Aquatic Insects* 13: 33–53.
- Stewart, K.W. and D.D. Zeigler. 1984. Drumming behavior of twelve North American stonefly (Plecoptera) species: First descriptions in Peltoperlidae, Taeniopterygidae and Chloroperlidae. *Aquatic Insects.* 6: 49–61.
- Watrous, L.E. and Q.D. Wheeler. 1981. The out-group comparison method of character analysis. *Syst. Zool.* 30: 1–11.
- Zeigler, D.D. and K.W. Stewart. 1977. Drumming behavior of eleven Nearctic stonefly (Plecoptera) species. *Ann. Entomol. Soc. Amer.* 70: 495–505.

Table 1. Drumming descriptions and signal characters for eight *Isoegenoides* species. Numbers of signals and individuals, numbers of beats per group, and overall beat intervals are provided for male calls, female answers, and male responses.

Species (Location Code)	No. Individuals		No. Signals		Range (Mode) Beats / Group		$\bar{x} \pm SD$ Beats / Group		Overall Beat intervals (msec) ($\bar{x} \pm SD$)		Overall Exchange intervals (msec) ($\bar{x} \pm SD$)		Overall Last σ - σ	
	δ	σ	δ	σ	δ	σ	δ	σ	δ	σ	δ - σ	σ - σ	δ - σ	Last σ - σ
<i>I. colubrinus</i>	115	114	113	3-7 (5)	1-4 (1)	1-6 (2)	65.0 \pm 8.3	121.8 \pm 26.8	441.8 \pm 142.5	402.0 \pm 101.2	1654.8 \pm 429.1	626.4 \pm 198.5		
(1) 5 σ 4 σ	5	4	5	5.2 \pm 0.7	1.8 \pm 0.8	2.5 \pm 0.9								
<i>I. doratus</i>	126	126		1-5 (3)	1-5 (2)		16.7 \pm 3.0	107.0 \pm 52.0		486.4 \pm 89.4		699.4 \pm 170.0		
(2) 1 σ 1 σ	1	1		3.1 \pm 0.6	2.1 \pm 1.0									
(3) 4 σ 6 σ	115	115		1-8 (3)	1-6 (1)		14.4 \pm 3.8	135.5 \pm 40.7		358.7 \pm 79.9		724.8 \pm 169.2		
	4	6		3.1 \pm 1.2	1.5 \pm 0.7									
<i>I. elongatus</i>	160	160	49	3-10 (7)	1-3 (3)	1-2 (1)	397.0 \pm 75.0	386.9 \pm 93.7	760.8 \pm 222.7	925.5 \pm 286.0	1971.9 \pm 159.8	888.8 \pm 294.8		
(4) 7 σ 3 σ	7	3	5	6.8 \pm 1.1	2.5 \pm 0.6	1.1 \pm 0.3								
(5) 4 σ 3 σ	121	120	57	4-8 (6)	1-3 (2)	1	222.1 \pm 14.0	180.5 \pm 20.7	N/A	570.0 \pm 82.1	950.2 \pm 124.1	648.3 \pm 178.6		
	4	3	4	5.9 \pm 0.7	1.8 \pm 0.5	1.0 \pm 0.0								
<i>I. frontalis</i> ,	135	135	132	2-6 (4)	1-3 (2)	1-8 (3)	61.2 \pm 11.3	93.8 \pm 29.3	288.8 \pm 129.4	561.5 \pm 76.3	1133.3 \pm 307.2	427.7 \pm 147.4		
(6) 7 σ 1 σ , (7) 1 σ ,	10	4	10	4.1 \pm 0.7	1.6 \pm 0.6	3.5 \pm 1.5								
(8) 3 σ 2 σ														
<i>I. hainsoni</i>	60			5-19 (6)			31.3 \pm 9.4							
(9) 4 σ	4			9.3 \pm 4.3										
<i>I. olivaceus</i>	269	243	88	5-8 (7)	1-10 (6)	1-6 (1)	98.2 \pm 12.0	86.6 \pm 22.0	356.5 \pm 144.1	739.9 \pm 130.9	831.6 \pm 231.1			
(10) 2 σ 1 σ ,	2	7	2	6.7 \pm 0.5	5.5 \pm 1.7	1.9 \pm 1.0								
(11) 6 σ														
<i>I. varians</i>	125	115	58	2-10 (6)	1-5 (4)	1-7 (3)	26.7 \pm 3.1	52.4 \pm 9.2	56.0 \pm 11.8	944.8 \pm 64.0	1285.7 \pm 276.3	892.8 \pm 200.1		
(12) 4 σ 5 σ	4	4	4	6.5 \pm 1.6	3.4 \pm 1.0	3.2 \pm 1.3								
(13) 4 σ 2 σ	117	100	38	2-11 (8)	1-5 (2)	1-5 (3)	36.9 \pm 4.0	56.7 \pm 9.7	73.4 \pm 21.4	839.9 \pm 61.4	1279.4 \pm 305.2	919.4 \pm 113.5		
	4	2	4	7.1 \pm 1.5	2.4 \pm 0.9	3.1 \pm 0.9								
<i>I. zionensis</i>	307	307	299	1-6 (4)	1-7 (2)	1-6 (3)	364.3 \pm 59.1	136.8 \pm 32.7	132.2 \pm 43.4	624.3 \pm 129.3	561.2 \pm 197.3	534.9 \pm 147.8		
(14) 7 σ 9 σ	7	9	7	4.0 \pm 0.6	2.2 \pm 1.1	3.7 \pm 1.4								
(14) 2 σ 5 σ	21	21	39	1-6 (4)	1-4 (2)	2-6 (4)	344.8 \pm 32.2	123.4 \pm 75.3	135.3 \pm 78.2	623.1 \pm 78.6	508.4 \pm 166.7	453.2 \pm 175.9		
	2	5	2	3.6 \pm 1.1	1.9 \pm 0.9	3.8 \pm 1.1								

†, (Sandberg and Stewart 2003)

Table 2. Drumming description and characters for *I. zionensis* at the colder 66° F (18.9° C). Numbers of signals and individuals and individuals, numbers of beats per group, and overall beat intervals are provided for male calls, female answers, or male responses.

Species (Location Code)	No. Signals		Range (Mode) Beats / Group		Overall Beat Intervals (msec) ($\bar{x} \pm SD$)		Overall Exchange Intervals (msec) ($\bar{x} \pm SD$)		Overall Last $\delta-\delta$		
	δ	δ	$\bar{x} \pm SD$	δ	δ	δ	δ	$\delta-\delta$	$\delta-\delta$	Last $\delta-\delta$	
<i>I. zionensis</i> (14) 2 δ 2 δ	138	115	124	2-5 (4)	1-5 (2)	589.2 \pm 50.7	206.7 \pm 54.9	184.7 \pm 37.9	2015.1 \pm 101.2	1327.0 \pm 171.3	921.2 \pm 321.8
(66° F)	2	2	2	4.2 \pm 0.5	2.3 \pm 1.1	3.7 \pm 1.1					

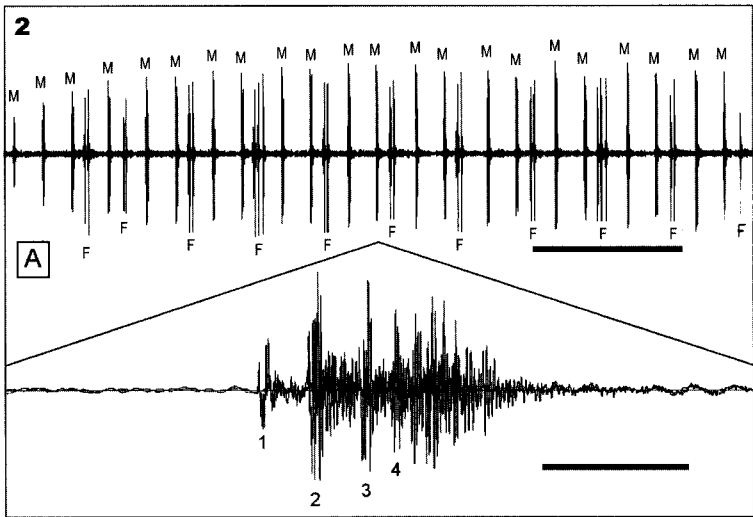
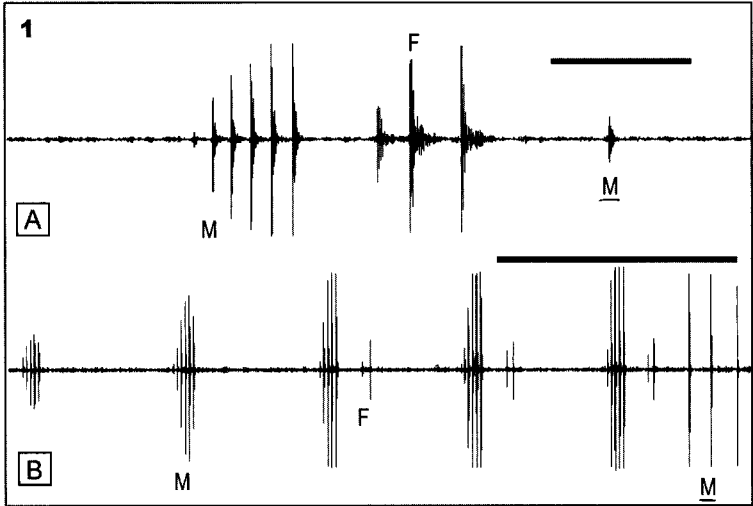


FIG. 1. *I. colubrinus* drumming. (A) Call-answer-response, 3-way sequence, Bar = 500 msec; (B) Grouped exchange. Bar = 5000 msec.

FIG. 2A. *Isogenoides doratus* drumming. Michigan Grouped exchange, Bar = 5000 ms, Total duration 24,288 msec. Dropdown: 100X horizontal zoom of male call group 12, Bar = 50 msec.

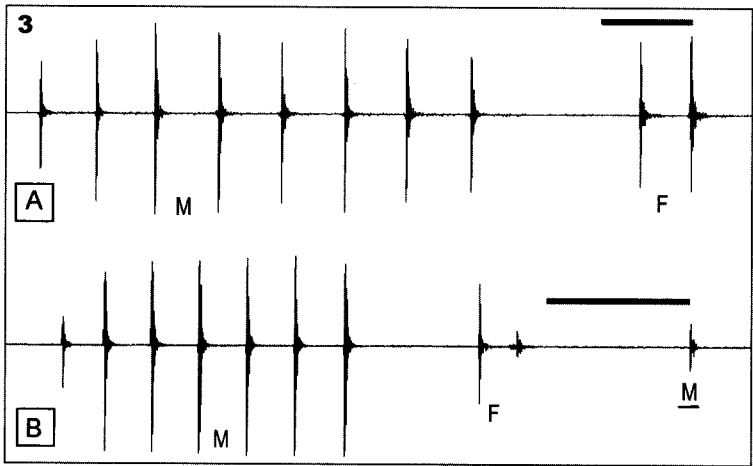
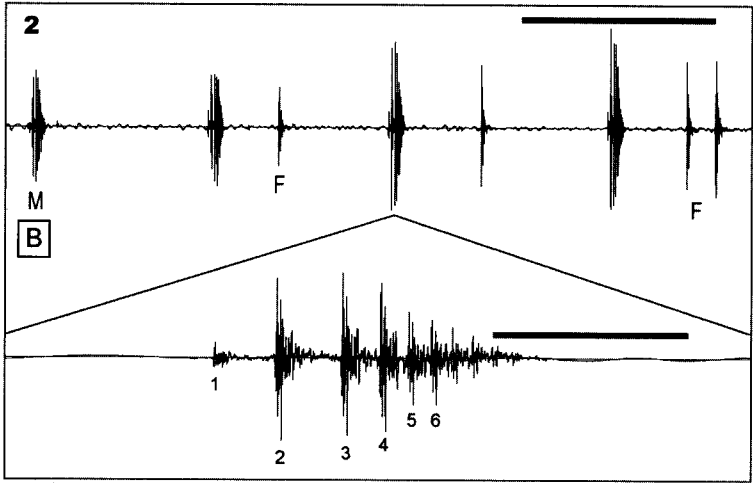


FIG. 2B. *I. doratus* drumming. Iowa Grouped exchange, Bar = 1000 msec. Dropdown: 20X horizontal zoom of male call group 3, Bar = 50 msec.

FIG. 3A, B. *I. elongatus* drumming. (A) 1999 Granby, CO, Sequenced call-answer, Bar = 500 msec. (B) 1999 Granby, CO, Call-answer-response, 3-way sequence, Bar = 1000 msec.

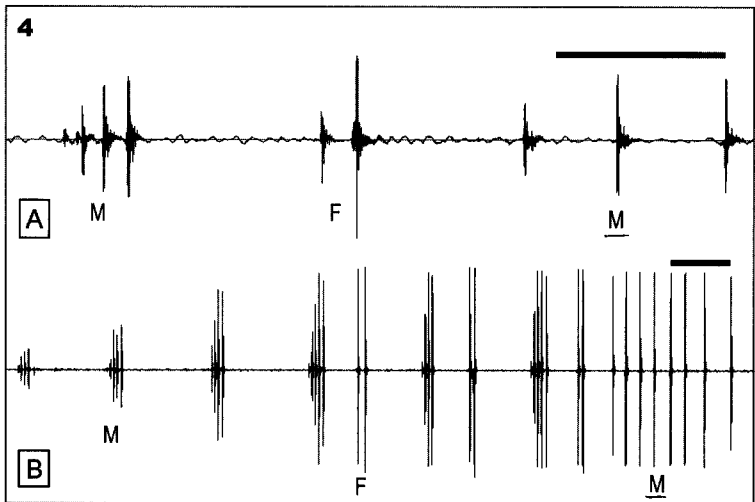
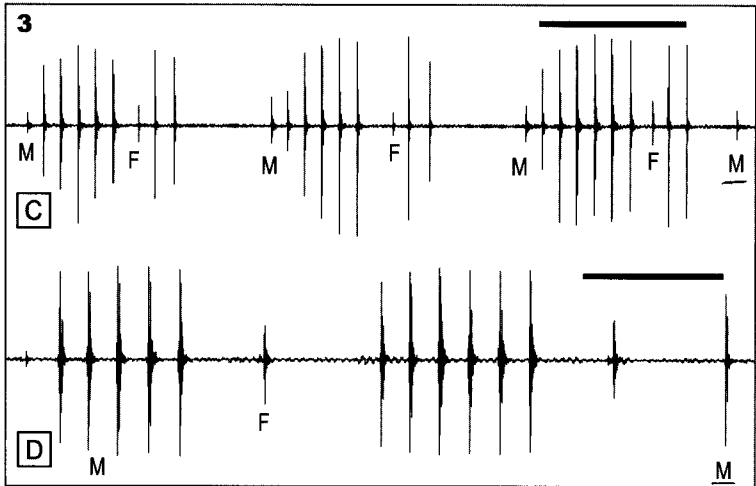


FIG. 3C,D. (C) *I. elongatus* drumming. 1999 Granby, CO, 7-way grouped exchange, Bar = 3000 msec. (D) 2000, Rifle, CO, 5-way grouped exchange, Bar = 1000 msec.

FIG. 4. *I. frontalis* drumming. (A) Call-answer-response, 3-way sequence, Bar = 500 msec. (B) Grouped exchange, Bar = 1000 msec.

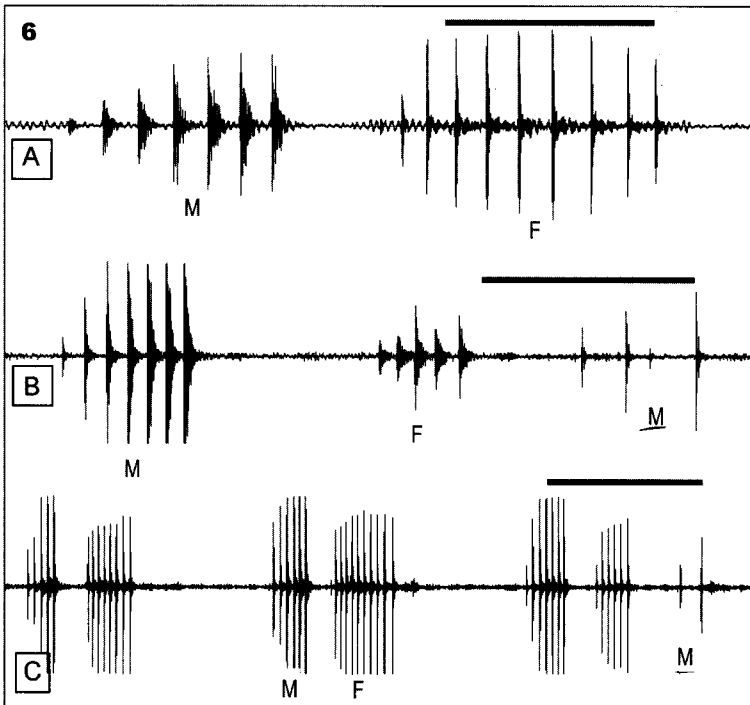
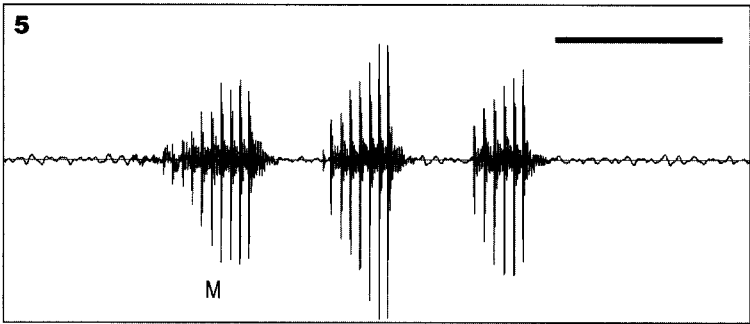


FIG. 5. *I. hansonii* drumming. Male grouped calls, Bar = 500 msec.

FIG. 6. *I. olivaceus* drumming. (A) Sequenced call-answer, Bar = 1000 msec. (B) Sequenced 3-way call-answer-response, Bar = 1000 msec. (C) Grouped 7-way exchange, Bar = 2000 msec.

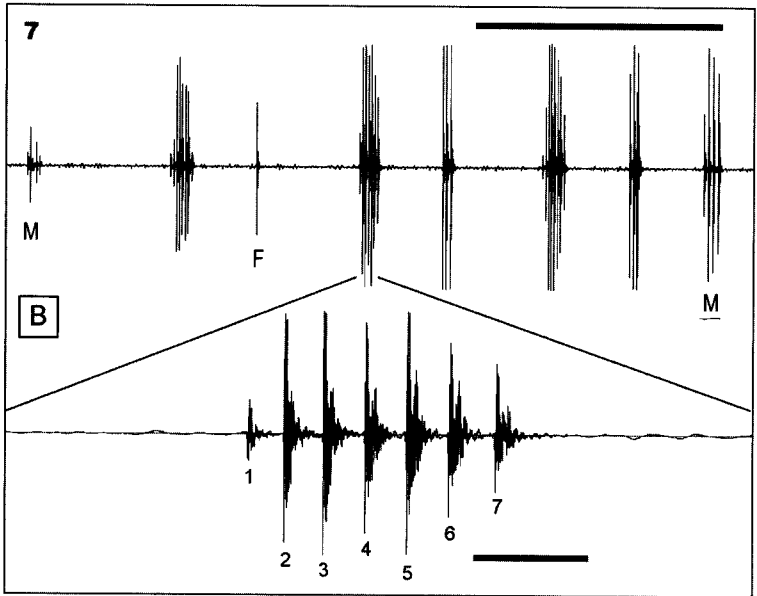
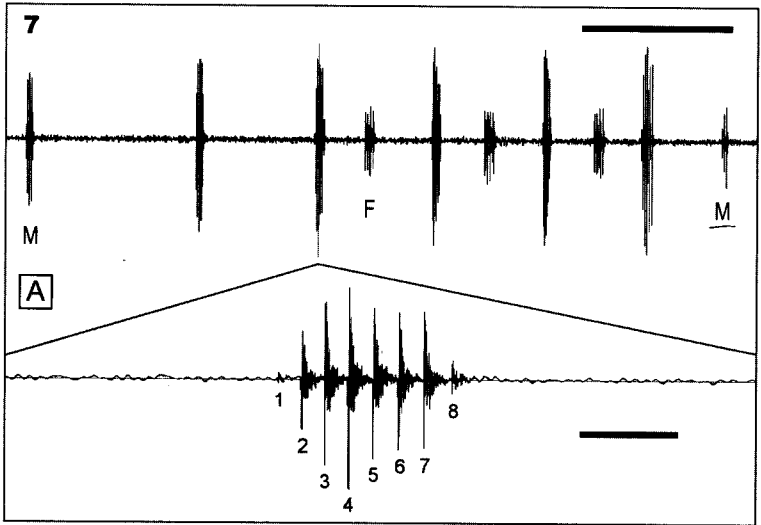


FIG. 7A. *I. varians* drumming. (A) 2000 Mississippi, grouped exchange, Bar = 3000 msec., Dropdown: 20X horizontal zoom of male call group 3, Bar = 100 msec.

FIG. 7B. 2001 Virginia, grouped exchange, Bar = 3000 msec., Dropdown: 15X horizontal zoom of male call group 3, Bar = 100 msec.

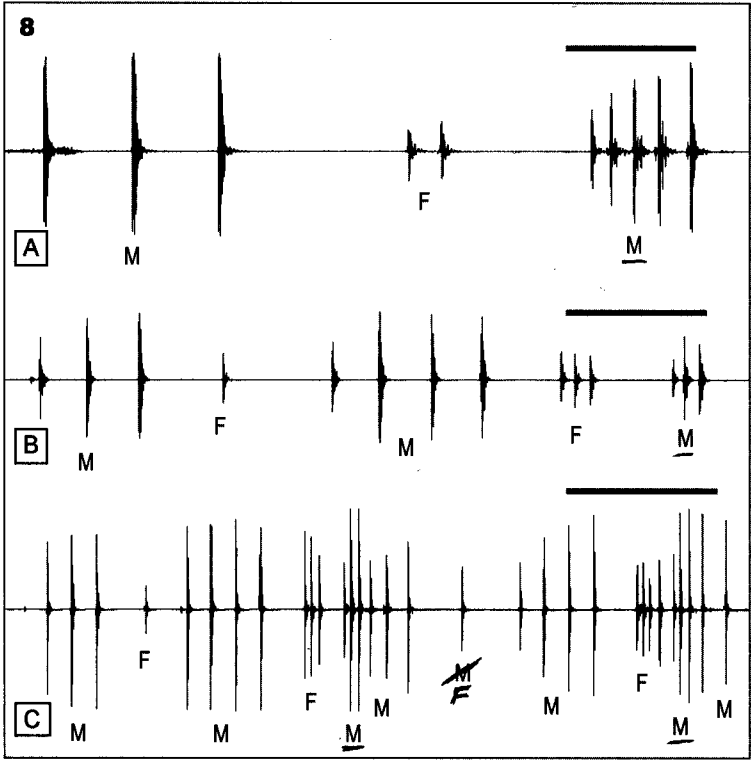


Figure 8. *I. zionensis* drumming. (A) Call-answer-response, 3-way sequence, Bar = 500 msec. (B) Grouped exchange, Bar = 1000 msec. (C) Complex grouped exchange, Bar = 2000 msec.

ERRATA

Vibrational Communication (Drumming) of the Nearctic Stonefly Genus *Isogenoides* (Plecoptera: Perlodidae)

Mistakenly the Acknowledgements section was not included in our submittal to the publisher. We submit them here.

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Corrections to text:

Page 116, Paragraph 6, Sentence 7:

“(...B-... B exchange interval)” should be (σ - σ exchange interval)

Page 118, “*Isogenoides zionensis*” heading should be italicized.

Page 120, Paragraph 6, Sentence 1:

“ ∞ ” symbol should be $^{\circ}$ (degree) symbol.

Page 130, Fig. 8-C:

“5th M” from left should be F (female)