SOUTHERN REGIONAL
AQUACULTURE CENTER

Annual Progress Report
May, 2014

DEVELOPMENT AND EVALUATION OF COOL-WATER CRAWFISH BAITS

Reporting Period
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Funding Level	Year 1	$37,595
	Year 2	$43,503
	Year 3	$43,796
	Total	$124,894

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PROJECT OBJECTIVES

1. Identify attractants, bait formulations, or potential manufacturing processes that increase the efficacy of formulated crawfish bait for use at water temperatures below 70 degrees F.

2. Compare the efficacy of experimental formulated baits or processes with fish baits for increasing crawfish catch and profits under simulated commercial conditions at water temperatures below 70 degrees F.

ANTICIPATED BENEFITS

Crawfish are harvested in over 185,000 acres of aquaculture ponds using baited wire-mesh traps that are lifted 3 to 5 days a week beginning as early as November and continuing through May to July of the following year. Traps are typically baited with manufactured formulated bait in warmer weather but, because formulated baits are inferior at cooler water temperatures, fresh-frozen cut fish is used. Fish for crawfish bait has become expensive, costing twice that of commercially formulated bait, and fish bait is frequently in short supply. More than half of the annual crawfish harvesting effort occurs during cool-water periods (December through late March), and with availability and price issues with fish, as well as the need to transport and store fish baits in a frozen state, this bait has become problematic for the crawfish industry. Development of an effective and economical cool-water formulated crawfish bait will address not only some of the cost and handling/storage issues with fish baits, but also will help conserve the fishery for many of these species.

PROGRESS AND PRINCIPAL ACCOMPLISHMENTS

Objective 1. *Identify attractants, bait formulations, or potential manufacturing processes that increase the efficacy of formulated crawfish bait for use at water temperatures below 70 degrees F.*

Louisiana State University Agricultural Center

A review of the scientific literature for identified attractants to carnivorous crustaceans revealed that amino acids and related biochemical compounds tend to elicit chemoattraction responses and may act as feeding stimulants for these crustaceans. Although some studies have shown that a feeding response can be elicited by single components, most have shown the response to be greatest with specific mixtures of amino acids or other compounds. Although detection does not equal attraction and an elicited feeding response may not equate to attraction over some distance to elicit entry of crawfish into a baited trap, this review provided creditable information.

Preliminary efforts were undertaken to establish an effective protocol for testing preferences of crawfish to attractants in the laboratory. Those efforts were not successful. The response of crawfish to field-proven attractants in a controlled laboratory environment, even at optimum temperatures with acclimated and/or starved captive stock, proved inconsistent and not predictable. Therefore, research was subsequently directed at developing a suitable technique “in the field” for effectively evaluating the efficacy of attractants in ponds that simulated commercial crawfish aquaculture. A gelatin-based matrix made without excessive heat or processing was found to be an effective medium to test attractants and could be used to evaluate potential attractants using commercial crawfish traps in experimental or commercial crawfish ponds. Several flesh-based attractants, including fish meal, when incorporated into the gelatin-based matrix, caught as many crawfish as cut gizzard shad (industry standard fish bait) when evaluated at temperatures from 51 to 63 degrees F. In contrast, concurrent field studies evaluated single amino acids, an amino acid mixture, sugar, fish oil and fish solubles incorporated into solid blocks of plaster of Paris (calcium sulfate dehydrate), as attractants for crawfish. The attractants imbedded in the plaster blocks were not effective when compared to either cut shad or attractants incorporated into the gelatin matrix. Thus, the gelatin matrix proved to be best suitable for identifying and testing potential attractants in experimental baits for crawfish in earthen ponds.

Initial tests during the first year of the project involved baits composed of the gelatin medium and various proteinaceous ingredients as test attractants. The test attractants consisted of selected commercially available ingredients, ground flesh products, and proprietary mixtures of synthetic amino acids. The experimental trials were conducted in either a commercial crawfish production pond or small research pond managed to simulate commercial crawfish ponds. Trials were conducted from January to early March at water temperatures ranging from 47 to 77 degrees F, and consisted of baited wire mesh traps (standard commercial trap of the crawfish aquaculture industry) placed at 45-feet intervals at random locations within the ponds. Trap soak duration was 24 hours. The response variables consisted of average number and weight of crawfish captured per trap per treatment. Capture rates with experimental baits were compared to cut fish (pogy, an industry standard), no bait, and in one trial, a commercially formulated bait (Purina Mills, Shreveport, LA). Results of these trials are provided in Table 1.

Of the experimental attractants tested, only minced pogy caught as many crawfish as cut pogy, the industry standard for cool water use. While other baits generally yielded higher catches than traps without bait, few differences were observed among the other proteinaceous attractants.

In year 2 of the project, additional ingredients were tested within the gelatin matrix for their relative effect on attracting crawfish to traps over five experimental trials. Catch results are presented in Table 2. Ingredients used for testing of attractant quality included several forms of fish products, various levels of fish product inclusion, a saccharide, an essential oil that exhibited potential for increasing catfish feed intake, and various commercially available high protein feed ingredients. Each trial also included treatments of cut pogy (menhaden), manufactured (warm-water) crawfish bait, and a non-baited control.

Results were mixed, with few test ingredients facilitating a catch rate nearing that of cut pogy. To determine if there were individual amino acids associated with better catch results, a correlation analysis was conducted. Individual amino acid concentrations (determined at Texas A&M University) for each test bait was correlated with the magnitude of that respective catch rate (standardized for the different trials by expressing catch in relation to cut pogy). Statistically significant correlation results are presented in Table 3.

Table 1. Average crawfish catch (by number and total weight of crawfish per trap), average weight of crawfish captured, and average catch (by number and by total weight) expressed as a percentage of that caught with cut menhaden (pogy) for experimental cool water attractants in 2011.

Treatment	No. trps	Avg. Catch (No/Trp)	Avg. Catch (Lb/Trp)	Wt. (g)	% of Cut Pogy (by No.)	% of Cut Pogy (by Wt.)
Trial 1 (temp = 47.2 min /56.7 max / 52.4 average)						
Cut frozen menhaden	3	4.8	0.20	17.4		
Gelatin-Minced Crawfish	3	0.5	0.02	15.0	10.4	10.0
Gelatin- Krill Meal	3	1.2	1.05	15.0	25.0	25.0
Gelatin- Squid Meal	3	2.3	0.07	14.1	47.9	35.0
Gelatin- Fish Meal	3	3.5	0.11	14.3	72.9	55.0
Gelatin- Minced Fresh Pogy	3	2.8	0.12	19.4	58.3	60.0
Gelatin- Minced Air-dried Pogy	3	5.5	0.22	17.6	114.6	110.0
No Bait	3	1.7	0.04	10.9	35.4	20.0
Trial 2 (temp = 53.2 min / 66.3 max / 60.2 average)						
Cut frozen menhaden	12	18.1	0.6	14.9		
Gelatin- Amino Acid Mix (1%)	12	10.3	0.31	13.5	56.9	51.7
Gelatin – No Attractant	12	9.3	0.32	15.5	51.4	53.3
Gelatin- Krill Meal	12	10.8	0.38	15.5	59.7	63.3
Gelatin- Squid Meal	12	11.1	0.38	14.8	61.3	63.3
Gelatin-Minced Crawfish	12	11.6	0.41	15.8	64.1	68.3
Gelatin- Fish Meal	12	13.6	0.47	15.9	75.1	78.3
Gelatin- Minced Fresh Pogy	12	21.8	0.69	14.3	120.4	115.0
Gelatin- Minced Air-dried Pogy	12	19.3	0.61	14.3	106.6	101.7
No Bait	12	5.3	0.16	13.8	29.3	26.7
Trial 3 (temp = 67.7 min / 76.7 max / 71.3 average)						
Cut frozen menhaden	12	24.9	0.96	17.6		
Gelatin- Amino Acid Mix (3%)	12	12.8	0.45	16.0	51.4	46.9
Gelatin-Minced Crawfish	12	15.8	0.62	17.9	63.5	64.6
Gelatin Fish/Squid/Krill Meal	12	16.3	0.62	17.1	65.5	64.6
Gelatin- Minced Fresh Pogy	12	26.2	0.99	17.2	105.2	103.1
Purina Pellets- Southern Pride	12	27.1	1.04	17.4	108.8	108.3
No Bait	12	6.3	0.2	14.3	25.3	20.8

During the last year of the project (year 3), single ingredients, high in one or more of the key amino acids identified in year 2 as desirable, and a custom mixture of synthetic amino acids were tested as attractants utilizing the gelatin-based bait matrix under commercial conditions. Capture results were compared with the industry standard (cut fish), as well as a popular warm water formulated bait (Purina) and a non-baited trap (Table 4).

Although subtle differences in catch were observed among the various attractants tested, the most notable observation was that poultry meal resulted in the best catch of all ingredients tested. Catch with Purina was unusually good in this trial compared to previous trials, likely a result of a high population density of crawfish in the pond. Poultry meal resulted in catches as good, or better, than Purina, and in several subsets exceeded that with cut fish. Poultry meal also was among the better test ingredients in a previous trial. Although poultry meal was not as effective as cut fish, it was consistent throughout the trial and was among the best single ingredients tested. It deserves further scrutiny for increased potential, possibly as a base for further formulation.

Table 2. Average crawfish catch (by number and total weight of crawfish per trap), average weight of crawfish captured, and average catch (by number and by total weight) expressed as a percentage of that caught with cut menhaden (pogy) for experimental cool water attractants in 2012. Values within columns, by trial, with the same superscript were not significantly different (P > 0.05). No significant differences were detected among treatments for mean size.

Treatment (Attractant)	Avg Catch (No/Trp)	Avg Catch (Lb/Trp)	Avg Wt. (g)	% of Cut Pogy (by No.)	% of Cut Pogy (by Wt.)
Trial 1: n=24 traps; Water Temperature = 56.8 min / 63.5 max / 61.0 average					
Cut frozen menhaden	27.6$^{\mathbf{A}}$	.89$^{\mathbf{A}}$	14.1	-	-
Catfish feed with EO*	7.4$^{\mathbf{BC}}$	.23$^{\mathbf{B}}$	13.7	26.8	25.8
EO (at 3%)*	6.5$^{\mathbf{BC}}$	.21$^{\mathbf{B}}$	14.2	23.6	23.6
EO (at 6%)*	5.8$^{\mathbf{BC}}$	.18$^{\mathbf{B}}$	13.9	21.0	20.2
Purina bait	12.2$^{\mathbf{B}}$	.37$^{\mathbf{B}}$	13.6	44.2	41.6
No bait	3.9$^{\mathbf{C}}$	.12$^{\mathbf{B}}$	13.8	14.1	13.5
Trial 2: n=16 traps; Water Temperature = 58.2 min / 62.5 max / 60.8 average					
Cut frozen menhaden	32.1$^{\mathbf{A}}$	1.23$^{\mathbf{A}}$	17.3	-	-
Freeze-dried menhaden meal (100%)*	27.3$^{\mathbf{A}}$	1.01$^{\mathbf{AB}}$	16.8	85.0	82.1
Freeze-dried menhaden meal (50%)*	26.2$^{\mathbf{A}}$	1.01$^{\mathbf{AB}}$	17.4	81.6	82.1
Freeze-dried menhaden meal (10%)*	14.8$^{\mathbf{B}}$	.59$^{\mathbf{C}}$	17.8	46.1	48.0
Freeze-dried menhaden meal (2%)*	12.4$^{\mathbf{B}}$	.47$^{\mathbf{CD}}$	16.7	38.6	38.2
Purina bait	18.6$^{\mathbf{B}}$	.70$^{\mathbf{BC}}$	17.1	57.9	56.9
No bait	5.3$^{\mathbf{C}}$	.19$^{\mathbf{D}}$	16.1	16.5	15.4
Trial 3: n=16 traps; Water Temperature = 60.3 min / 64.1 max / 62.6 average					
Cut frozen menhaden	35.8$^{\mathbf{B}}$	1.45$^{\mathbf{AB}}$	18.2	-	-
Minced fresh fish*	51.2$^{\mathbf{A}}$	2.1$^{\mathbf{A}}$	18.4	143.0	144.8
Solvent extracted freeze-dried menhaden meal*	39.6$^{\mathbf{AB}}$	1.61$^{\mathbf{AB}}$	18.1	110.6	111.0
Freeze-dried menhaden meal*	32.3$^{\mathbf{B}}$	1.39$^{\mathbf{B}}$	19.4	90.2	95.9
Freeze-dried menhaden meal (heated)*[1]	32.9$^{\mathbf{B}}$	1.36$^{\mathbf{B}}$	18.6	91.9	93.8
Minced oven dried menhaden (low temp)*[2]	32.9$^{\mathbf{B}}$	1.37$^{\mathbf{B}}$	18.8	91.9	94.5
Minced oven dried menhaden (high temp)*[1]	24.9$^{\mathbf{BC}}$	1.04$^{\mathbf{BC}}$	18.7	69.6	71.7
Purina bait	31.6$^{\mathbf{B}}$	1.31$^{\mathbf{B}}$	18.8	88.3	90.3
No bait	11.5$^{\mathbf{C}}$	.42$^{\mathbf{C}}$	16.5	32.1	29.0
Trial 4: n=16 traps; Water Temperature = 52.8 min / 58.2 max / 55.9 average					
Cut frozen menhaden	50.9$^{\mathbf{A}}$	2.04$^{\mathbf{A}}$	18.2	-	-
Poultry by-products*	32.1$^{\mathbf{B}}$	1.04$^{\mathbf{B}}$	18.9	63.1	51.0
Dried grains w/solubles*	21.9$^{\mathbf{CD}}$	.92$^{\mathbf{B}}$	19.0	43.0	45.1
Fish meal 200%*	21.9$^{\mathbf{CD}}$	.92$^{\mathbf{B}}$	19.2	43.0	45.1
Fish meal 100%*	19.6$^{\mathbf{CD}}$	.81$^{\mathbf{BC}}$	19.0	38.5	39.7
Fish meal/soybean meal*	16.0$^{\mathbf{D}}$	.63$^{\mathbf{C}}$	17.8	31.4	30.9
Soy protein concentrate*	15.0$^{\mathbf{D}}$	.62$^{\mathbf{C}}$	19.0	29.5	30.4
Soybean meal in matrix*	13.8$^{\mathbf{DE}}$	.54$^{\mathbf{C}}$	17.9	27.1	26.5
Purina bait	26.6$^{\mathbf{BC}}$	1.05$^{\mathbf{B}}$	17.9	52.3	51.5
No bait	5.0$^{\mathbf{E}}$	.20$^{\mathbf{D}}$	17.9	9.8	9.8
Trial 5: n=14 traps; Water Temperature = 56.7 min / 61.7 max / 59.6 average					
Cut frozen menhaden	25.9$^{\mathbf{A}}$	.94$^{\mathbf{A}}$	17.7	-	-
Fish meal*	15.5$^{\mathbf{B}}$	.54$^{\mathbf{B}}$	17.0	59.8	57.4
Fish meal + sugar (20%)*	13.8$^{\mathbf{BC}}$	.52$^{\mathbf{B}}$	18.1	53.3	55.3
Sugar*	10.7$^{\mathbf{C}}$	.38$^{\mathbf{BC}}$	17.1	41.3	40.4
Purina bait	26.2$^{\mathbf{A}}$	.96$^{\mathbf{A}}$	17.7	101.2	102.1
No bait	5.9$^{\mathbf{D}}$	.22$^{\mathbf{C}}$	17.0	22.8	23.4

*Indicates attractant was contained within the gelatin matrix

[1] Drying/heating temperature = 90 C.

[2] Drying temperature = 60 C.

Table 3. Correlation coefficients (r value) for selected amino acid levels in test baits and corresponding relative catch values by number of crawfish caught per trap for those baits. Amino acid (AA) levels in cut menhaden (pogy) as well as freeze-dried pogy (***Best*** showing category of test baits), fish meal (***Mediocre*** showing category), and soybean meal (***Poor*** showing category) are also included. Amino acid concentrations expressed as nmol/mg of wet weight.

AA (or Derivative)	r value	P value	AA level in Cut Pogy	AA level in FD Pogy	AA level in Fish Meal	AA level in Soy Meal
Serine	.88897	<.0001	1.69	5.52	2.52	.16
Tyrosine	.87619	<.0001	0.58	3.08	1.62	.13
Threonine	.87169	.0001	0.77	3.18	2.38	.10
Aspartate	.87139	.0001	0.07	2.88	1.34	1.24
Glutamine	.82951	.0005	0.65	5.06	1.02	0
Lysine	.82459	.0005	0.91	4.64	3.38	.17
Histidine	.81087	.0008	5.27	12.16	9.44	.21
Cystathionine	.78869	.0014	0.04	.19	.09	.03
Phenylalanine	.77874	.0017	0.71	2.57	2.60	.26
1-Methylhistidine	.76106	.0025	0.07	.32	.27	.08
Cystine	.76051	.0025	0.04	.25	.03	.02
Leucine	.72484	.0051	1.45	4.83	6.38	.12
Taurine	.72445	.0051	8.46	18.14	24.43	.01
Isoleucine	.69781	.0080	0.57	2.30	3.33	.12
Glutamate	.6815	.0103	0.84	5.06	2.19	1.0
Valine	.67707	.0110	1.02	3.55	5.51	.19
3-Methylhistidine	.66849	.0125	0.18	1.05	.86	.11
Glycine	.65795	.0145	1.34	4.34	5.38	.27
Methionine	.64992	.0162	0.33	.38	.72	.09
Alanine	.57872	.0382	2.86	9.42	12.82	.77

Auburn University

Dr. Allen Davis has provided technical support for the project. This has included direction of the project, recommendations for sources and types of test ingredients and has provided insight into possible manufacturing processes. Additionally, he has provided several ingredients for testing. He will assist in reviewing and analyzing research results in their totality.

Objective 2. *Compare the efficacy of experimental formulated baits or processes with fish baits for increasing crawfish catch and profits under simulated commercial conditions at water temperatures below 70 degrees F.*

Louisiana State University Agricultural Center

While all tests conducted involved a comparison with cut fish (the current industry standard for cool water) as the attractant, capture rate of non-baited traps is also included for every experimental trial. Because a suitable alternative to cut fish was not identified within the limited budget and time frame of this project, efficacy and cost evaluations of a formulated product or different processing methods were not possible.

Table 4. Summary of results for experimental trials during Year 3 of the cool-water crawfish bait study. No significant differences were detected among treatments for mean weight at harvest. Catch, both by number of crawfish and weight of crawfish by trap, is also expressed as a percentage of that caught with cut pogy (Fish).

Treatment (Attractant)[1]	Avg Catch (No/Trp)	Avg Catch (Lb/Trp)	Avg Wt. (g)	% of Cut Menhaden (by No.)	% of Cut Menhaden (by Wt.)
n=20 traps; Water Temperature(F) = 53 min / 64 max / 58 average					
No Bait	6.2^{G}	0.26^{G}	19.3	21.5	20.3
Guano	8.8^{FG}	0.34^{G}	17.4	30.3	26.7
Fmeal++	9.6^{EFG}	0.44^{FG}	21.3	33.3	34.8
Fmeal	10.6^{DEFG}	0.42^{FG}	18.7	36.7	32.6
Fmeal+G	10.6^{DEFG}	0.42^{FG}	17.4	36.7	33.2
Bmeal	10.7^{DEFG}	0.44^{FG}	18.9	37.1	34.5
Fmeal+T	10.9^{DEFG}	0.43^{FG}	18.4	37.6	33.5
Fhydro	11.3^{DEFG}	0.52^{EFG}	20.9	39.0	40.5
AA2x	15.5^{CDEF}	0.67^{DEF}	20.2	53.7	52.2
Grobio	16.5^{BCDE}	0.70^{CDEF}	18.5	57.2	54.5
AA	16.7^{BCDE}	0.70^{CDEF}	19.0	57.9	54.8
Crab	17.4^{BCD}	0.77^{CDE}	20.7	60.1	60.4
Pmeal-R	22.4^{ABC}	1.05^{AB}	21.8	77.6	82.3
Purina	22.6^{ABC}	0.87^{BCD}	18.3	78.2	67.9
Pmeal-P	22.8^{AB}	0.95^{BC}	19.5	78.9	74.8
Fish	28.9^{A}	1.28^{A}	20.8	-	-

[1] Treatments were no bait, seabird guano (Guano), feather meal plus taurine and glycine (Fmeal++), feather meal (Fmeal), feather meal plus glycine (Fmeal+G), bloodmeal (Bmeal), feather meal plus taurine (Fmeal+T), fish hydrolyzate (Fhydro), amino acid mixture 2x rate (AA2X), Grobiotic-A (Grobio), amino acid mixture (AA), crab meal (Crab), poultry meal – regular ash (Pmeal-R), Purina manufactured crawfish bait (Purina), poultry meal – pet food grade (Pmeal-P), and fish (cut pogy).

Texas A&M University

A total of 16 different test baits for crawfish were analyzed in year 2 for amino acid composition of their protein-bound and free-pool constituents. For most baits (Essential oil had none), 23 primary and 18 secondary amino acids were detected in the free pool; whereas, 18 amino acids were detected in the protein-bound form. Regression analysis showed that 23 amino acids significantly correlated with catch values obtained in earlier trials at Louisiana State University. Only two amino acids correlated with catch were in the protein-bound form and only seven had correlation values above 0.8 (all free amino acids) (Table 3).

In addition, the correlations seem to point to a limited number of amino acids that are found in relatively high concentrations in the fresh pogy and the other highest ranking baits. These include taurine, histidine, alanine, lysine, serine, leucine, glycine, valine, and glutamate (Table 3). On the other hand, most of these amino acids were in lower concentrations in the mediocre and poor ranking baits than in fresh pogy. Nevertheless, the finding of two mediocre showing baits, fish and poultry meals, having higher absolute values of all these amino acids than the fresh pogy, seems to point out that other non-detected attractant(s) may be present in the latter bait, ergo not present in the former two meals.

Based on these findings, a follow-up study was carried out with the fresh pogy – control bait with excellent showing – and the Purina bait – mediocre showing – to identify top-leaching amino acids from soaked baits throughout an 8-h period. Results from this assay showed five prominent amino acids in water containing fresh pogy, which in the order of concentration were histidine, taurine, alanine, lysine and glycine (Fig. 1); however, histidine and taurine were in the range of 4- to 6-fold higher than the other three amino acids. On the other hand, the top five amino acids in water containing the Purina bait were glutamate, aspartate, taurine, alanine, and glycine (Fig. 2). Interestingly, although both baits had similar amino acids within the top five (e.g., taurine, alanine and glycine), the release of them to the water was much slower in soaked Purina bait than in fresh pogy.

Figure 1. Top five amino acid concentrations (nmol/mL) in water containing fresh pogy at different bait:water ratios, 1:4 (A) and 1:16 (B). Histidine and taurine were 4- to 6- fold higher in both ratios compared to alanine, lysine and glycine.

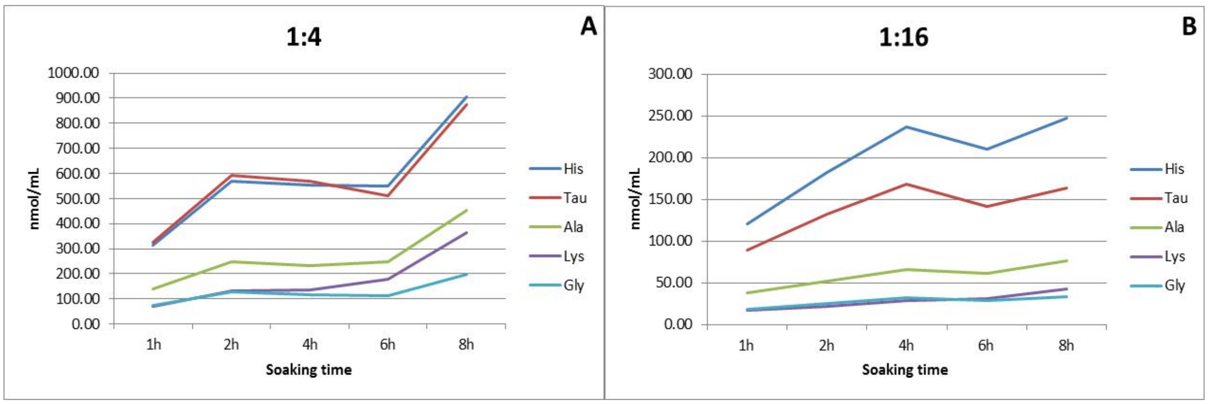
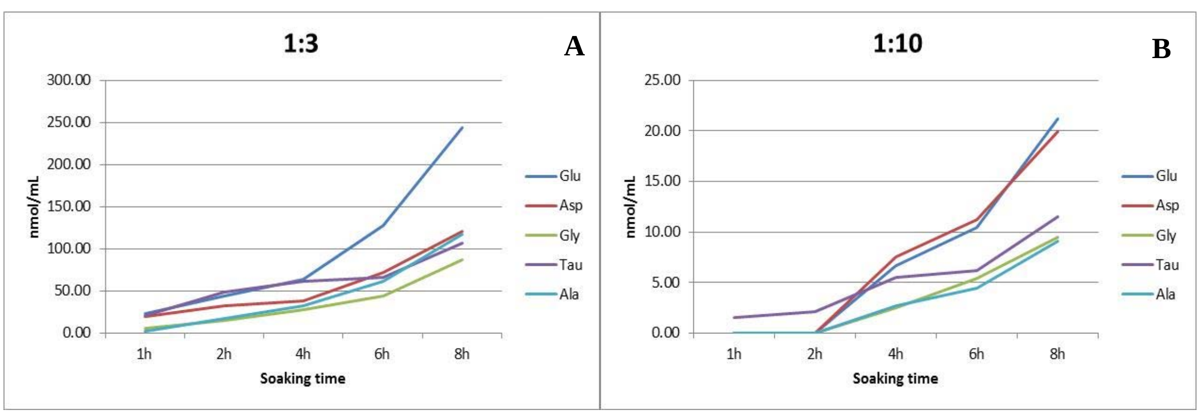


Figure 2. Top five amino acid concentrations (nmol/mL) in water containing Purina bait at different bait:water ratios, 1:3 (A) and 1:10 (B). Amino acid release was slower with this bait, and at the end of the assay glutamate was 2-fold higher than the other amino acids at 1:3 ratio; whereas, at 1:10 glutamate and aspartate were 2-fold higher than glycine, taurine and alanine.



IMPACTS

The primary impact of the results to date has been to provide scientists with valuable information regarding what direction to take future research in an attempt to identify methods and potential attractants for further testing. The recent findings have provided quantitative assessments of the value of limited substances and feedstuffs as potential crawfish attractants, and have provided valuable information regarding certain physical aspects needed for effective crawfish baits. Specifically, this research suggests that key amino acids may play a vital role in determining the quality of a crawfish attractant and have identified several amino acids that may be most important. Moreover, it was determined that the rate and timings of key amino acid released from baits in water may affect the efficacy of different baits. This provides the impetus and possible direction for further research.

PUBLICATIONS, MANUSCRIPTS OR PAPERS PRESENTED

McClain, W.R. and J.J. Sonnier. 2011. Investigation of test ingredients as potential attractants for crawfish in cool water. Ann. Res. Rpt., Rice Res. Stn., La. Agri. Exp. Stn., L.S.U. Agricultural Center, 103:257-262.

McClain, W.R., D. Gatlin, III, and J.J. Sonnier. 2012. Investigation of test ingredients as attractants for crawfish in cool water. Ann. Res. Rpt., Rice Res. Stn., La. Agri. Exp. Stn., L.S.U. Agricultural Center, 104:286-302.

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