CHEMORECEPTION IN THE BUCCINID GASTROPODS, *BABYLONIA ZEYLONICA* AND *BABYLONIA SPIRATA* (NEOGASTROPODA : BUCCINIDAE)

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ABSTRACT

The proboscis search reaction in *Babylonia zeylonica* and *Babylonia spirata* was determined using biological extracts, organic and inorganic compounds. Twenty snails were used for each test. Extracts of six species of marine animals were tested: oyster *Crassostrea madrasensis*, clam *Meretrix meretrix*, squid *Loligo* sp., shrimp *Penaeus indicus*, crab *Portunus pelagicus*, and fish *Leognathus* sp. Seven organic compounds were tested: egg albumin flake, sucrose, maltose, dextrose, lactose, starch and glycogen. The responses to 3 acids, 3 bases, and 10 salts were tested. Positive responses were obtained with extracts of *C. madrasensis*, *P. indicus*, *M. meretrix*, pure protein, carbohydrates and salts, while escape responses were recorded with acids and bases.

INTRODUCTION

The physiological process which occurs in the receptor cells upon chemical stimulation is termed chemoreception (Hodgson 1964). Chemoreception triggers a wide variety of behavioural patterns: identification of potential food sources, feeding initiation, detection of predators, noxious stimuli, sexual identification, and synchronized spawning (Audesirk & Audesirk 1985). The present study was designed to investigate positive or negative responses in terms of proboscis search reactions. The carnivore snails *Babylonia zeylonica* and *Babylonia spirata* were tested in the laboratory with organic and inorganic compounds.

MATERIALS AND METHODS

Materials: The snails were collected at Porto Novo (Lat. 11°29'N, Long. 79°47'E), Bay of Bengal, using traps at 6-10 m depth. The snails were acclimatized, and fed with bivalves for one month, before the tests. *B. zeylonica* were 6.7-7.3 cm (total length) and 32.3-39.4 g (total weight). *B. spirata*. were 5.6-6.1 cm (total length) and 35.5 - 45.3 g (total weight).

Biological extracts were prepared from fresh oyster, *Crassostrea madrasensis*, clam *Meretrix meretrix*, squid *Loligo* sp., shrimp *Penaeus indicus*, crab *Portunus pelagicus* and fish *Leognathus* sp. The muscular parts were homogenized in filtered sea water 1:1 (v/w). From the crude extract, 10 samples at 10-100 % concentrations were prepared by dilution with filtered sea water.

Organic compounds. Solutions of protein and carbohydrate were prepared separately in filtered sea water. 10 samples at 1-10 % concentrations were made of egg albumin flake, sucrose, maltose, dextrose, lactose, starch and glycogen.

Inorganic compounds. Acids, bases and salts were used. Ten concentrations (1-10 %) of each acid and base were prepared with filtered sea water. Salts were prepared at 20 concentrations (1-20 %) with filtered sea water. The following compounds were tested. Acids: HCl, HNO_3, H_2SO_4. Bases: KOH, NaOH, NH_4OH. Salts: $MgCl_2$, NaCl, $CaCl_2$, $CuSO_4$, KCl, KBr, KI, $NaNO_3$, NH_4Cl.

Blotting. Biological extracts, protein, and carbohydrate solutions (2 ml) were dropped on Whatman No.1 filter paper, 5 cm diameter. The soaked filter papers were air dried and stored in a desiccator.

Experimental design. Twenty snails of each species were used at each concentration. Experiments were conducted in circular plastic tanks (50 cm diameter). Filtered sea water (10 l) was used for each trial. Before an experiment, the snails were starved for two days, then placed at the margin of the tank, and the blotted filter paper put in the centre. Fifteen minutes were given to each snail to perceive the stimuli emanating from the filter paper. The number of snails moving towards the stimulus, the time to react, and the time to reach the stimulus, were recorded.

The response to inorganic compounds was determined by dropping a test solution near the siphon of active snails. A 50 ml burette with a flexible polyethylene tube (0.5 cm diameter) was used. The behaviour of twenty individuals of each species were tested, and the response recorded at each concentration.

RESULTS

B. zeylonica and *B. spirata* could quickly perceive the substances emanating from extracts on the filter paper. Once the snails were stimulated to feed, they extended their proboscis to a variable degree in search for food. The snails could perceive the stimulatory substances, as well as reach the stimuli quickly for both biological extracts and organic compounds. The proboscis was extended when a substance stimulated feeding. This behaviour is referred to as a positive response. If a snail disliked a given stimulus, it would withdraw into the shell or try to get away from the stimulus. The proboscis was never extended. This behaviour is termed escape response. The osphradium is considered to be a chemoreceptive organ in gastropods and the snails perceive the stimuli through the respiratory current. The time needed for olfactory detection may vary between species. Therefore, the perception time as well as the time to reach the stimuli were recorded for the two species of *Babylonia*.

Biological extracts. Tables 1-6 show the mean time spent for perception, and the time to reach the stimuli. The biological extracts stimulated feeding response in the following order: oyster > shrimp > clam > squid > fish > crab.

Organic compounds. Both species displayed positive feeding response at the protein concentrations tested (Table 7). The 6 carbohydrates stimulated feeding in the following order: glycogen > sucrose > lactose > maltose > dextrose > starch (Tables 8-13).

Inorganic compounds. All tested acids and bases resulted in 100 % escape response, even at the lowest concentration, in both the species, although *B. spirata* seemed to be slightly more tolerant than *B. zeylonica*.. The salts lithium chloride, magnesium chloride, and sodium chloride resulted in positive feeding response in both species. Other salts had no effect, or they resulted in negative response with increasing concentrations (Table 14).

DISCUSSION

All the tested biological extracts stimulated extension of the proboscis. A 100 % feeding response was observed at higher concentrations, whereas the percentage of response was lower at the lower concentrations. Hara (1982) found that the positive response to crude biological extracts seems to involve the combined effect of several compounds, because the response is greater compared to pure substances. Gurin & Carr (1971) found that oyster fluid contained more than one major protein component. The concentrations which stimulated a 50 % positive response in *Nassarius obsoletus* were approximately 1 to 2×10^{-10} molar. Carr (1967) identified the most obvious group of stimulatory compounds in shrimp extracts. They were amino acids, especially glycine, betaine, and lactate.The response inducing compounds obtained from shrimp extracts were substances of low molecular weight (Carr *et al.* 1974). But in many cases, compounds of relatively high molecular weight are important stimulants, *e.g.*, in *Nassarius obsoletus*. Gurin and Carr (1971) found that major response inducing capacity was attributed to specific proteins. Solms (1969) stated that the macromolecules such as proteins can provide a high degree of specificity for the transformation of chemical information. Lindstedt (1971) reported that glycoprotein and albumin were feeding stimulants in *Nassarius* sp.

Carbohydrates stimulated feeding responses in both *B. zeylonica* and *B. spirata*, but only at higher concentrations (10 %), except glycogen which gave a 100 % feeding response at 3 %. When compared to protein, perception time, and the time to reach the stimuli were longer for carbohydrates. Henschel (1932) found that glycogen, starch and sucrose stimulated feeding responses in *Nassarius reticulata* at concentrations of 0.03-2.5 g/l, 0.25-40 g/l and 0.5-10 g/l respectively. Jager (1971) found that the disaccharides sucrose, maltose, and lactose were more stimulatory than the monosaccharides D-glucose, D-fructose, and D-galactose in *Lymnaea stagnalis*. The above concentrations of carbohydrates, resulting in feeding response of other gastropods, are comparable to the present concentrations.

Acids and bases induced escape response in *Babylonia*. This was to be expected. Hoffmann (1930) observed a defensive responses in *Nassarius mutabilis* exposed to HCl, HNO_3 and H_2So_4 at concentrations of 1.25-10 g/l, 0.75-30 g/l and 0.05-10 g/l respectively, and to the bases potassium hydroxide at 1.0-100 g/l, sodium hydroxide 0.5-75 g/l, and ammonium hydroxide 8.75 g/l. Wrede (1928) reported that *Physa fontinalis* responded defensively to KOH at concentrations of 0.5-1 g/l and to NaOH at 0.2-1 g/l.

Among the salts tested, lithium chloride, magnesium chloride and sodium chloride gave a positive response in both species. However, the required concentrations varied among the salts. In *Nodilittorina*, Ohasawa & Tsukuda (1955) found a positive response with NaCl+$MgCl_2$ while the response was slight when NaCl was tested alone. In comparison, $MgCl_2$ alone was effective in about the half of the trials. It suggests that Mg^{++} is the primary stimulus, as also found in the present study. It is speculated that the proboscis search reaction is a response to salts which may leak from the food source in nature.

Table 1. Perception and feeding response stimulation of oyster extract in A = concentration (%); *B. zeylonica*: B = Time spent for the perception; C = Time to reach the stimulus; D = responding snails (%); *B. spirata*: E = Time spent for the perception; F = Time to reach the stimulus. Times in seconds, calculated as the mean of maximal and minimal values (n = 20).

A (%)	B (sec)	C (sec)	D (%)	E (sec)	F (sec)	G (%)
10	36	154	35	44	190	30
20	32	165	65	35	172	50
30	29	133	85	29	161	75
40	24	124	100	25	146	100
50	22	121	100	23	127	100
60	19	128	100	21	89	100
70	18	109	100	18	104	100
80	15	98	100	16	111	100
90	12	83	100	16	113	100
100	10	75	100	13	80	100

Table 2. Perception and feeding response stimulation of clam extract. *see* legend explanation in text to Table 1.

A (%)	B (sec)	C (sec)	D (%)	E (sec)	F (sec)	G (%)
10	59	213	25	86	218	20
20	50	201	35	75	209	30
30	43	189	55	61	192	50
40	40	177	75	49	178	70
50	34	161	90	43	159	85
60	28	152	100	38	150	100
70	25	139	100	30	144	100
80	22	128	100	24	136	100
90	17	116	100	20	121	100
100	15	98	100	17	108	100

Table 3. Perception and feeding response stimulation of squid extract. *see* legend explanation in text to Table 1.

A (%)	B (sec)	C (sec)	D (%)	E (sec)	F (sec)	G (%)
10	160	270	10	170	291	5
20	146	255	15	156	270	10
30	129	245	30	137	257	20
40	117	233	45	120	241	40
50	102	224	65	106	233	60
60	87	214	85	87	220	75
70	72	206	100	76	212	85
80	59	197	100	62	201	100
90	48	189	100	54	194	100
100	40	174	100	43	183	100

Table 4. Perception and feeding response stimulation of shrimp extract. *see* legend explanation in text to Table 1.

A (%)	B (sec)	C (sec)	D (%)	E (sec)	F (sec)	G (%)
10	82	263	40	81	269	30
20	72	241	55	73	253	50
30	63	228	75	67	233	70
40	46	215	90	59	220	85
50	40	196	100	53	199	100
60	35	182	100	45	184	100
70	30	169	100	39	169	100
80	25	154	100	32	152	100
90	21	143	100	26	146	100
100	18	135	100	23	141	100

Table 5. Perception and feeding response stimulation of crab extract. *see* legend explanation in text to Table 1.

A (%)	B (sec)	C (sec)	D (%)	E (sec)	F (sec)	G (%)
10	-	-	-	-	-	-
20	-	-	-	-	-	-
30	194	332	5	-	-	-
40	182	319	15	187	322	5
50	165	305	30	166	307	20
60	153	290	40	153	291	35
70	142	277	55	139	280	45
80	121	271	75	126	268	60
90	108	246	95	114	259	85
100	96	246	100	103	254	100

Table 6. Perception and feeding response stimulation of fish extract. *see* legend explanation in text to Table 1.

A (%)	B (sec)	C (sec)	D (%)	E (sec)	F (sec)	G (%)
10	-	-	-	-	-	-
20	153	295	10	-	-	-
30	135	278	25	154	286	15
40	126	265	35	142	270	25
50	117	255	50	129	260	45
60	102	200	65	119	248	60
70	92	224	85	103	239	75
80	85	212	100	93	221	80
90	75	203	100	88	210	100
100	62	194	100	78	201	100

Table 7. Perception and feeding response stimulation of protein. *see* legend explanation in text to Table 1.

A (%)	B (sec)	C (sec)	D (%)	E (sec)	F (sec)	G (%)
1	35	143	90	36	147	80
2	31	131	100	33	136	100
3	29	126	100	31	129	100
4	25	116	100	28	117	100
5	22	108	100	25	107	100
6	19	98	100	23	94	100
7	15	88	100	17	83	100
8	13	82	100	14	78	100
9	9	73	100	11	74	100
10	7	68	100	8	71	100

Table 8. Perception and feeding response stimulation of sucrose. *see* legend explanation in text to Table 1.

A (%)	B (sec)	C (sec)	D (%)	E (sec)	F (sec)	G (%)
1	160	305	20	170	317	15
2	149	282	35	154	293	30
3	140	275	50	143	281	50
4	129	257	65	135	260	50
5	120	248	70	124	249	65
6	104	231	80	107	240	75
7	95	224	80	97	229	80
8	82	215	90	88	219	90
9	75	208	95	78	212	90
10	64	198	100	68	201	100

Table 9. Perception and feeding response stimulation of maltose. *see* legend explanation in text to Table 1.

A (%)	B (sec)	C (sec)	D (%)	E (sec)	F (sec)	G (%)
1	-	-	-	-	-	-
2	178	336	20	185	340	10
3	160	312	30	167	309	25
4	141	300	50	145	289	45
5	127	288	65	130	281	65
6	109	274	70	116	274	75
7	98	266	85	103	267	75
8	82	257	85	85	264	80
9	75	245	90	79	257	90
10	69	183	100	73	245	100

Table 10. Perception and feeding response stimulation of dextrose. *see* legend explanation in text to Table 1.

A (%)	B (sec)	C (sec)	D (%)	E (sec)	F (sec)	G (%)
1	-	-	-	-	-	-
2	191	339	20	-	-	-
3	168	327	35	174	339	25
4	148	314	50	158	323	40
5	131	303	50	142	305	50
6	118	281	65	123	282	65
7	102	267	80	106	276	75
8	90	263	90	92	267	80
9	80	256	90	89	261	90
10	70	245	100	81	253	100

Table 11. Perception and feeding response stimulation of lactose. *see* legend explanation in text to Table 1.

A (%)	B (sec)	C (sec)	D (%)	E (sec)	F (sec)	G (%)
1	164	299	20	176	305	10
2	152	287	30	157	294	25
3	141	278	45	145	283	40
4	128	265	60	134	269	55
5	115	252	60	118	256	60
6	102	243	75	105	245	70
7	93	229	80	97	233	70
8	83	220	90	84	224	85
9	77	214	90	80	214	90
10	66	206	100	68	210	100

Table 12. Perception and feeding response stimulation of starch solution. *see* legend explanation in text to Table 1.

A (%)	B (sec)	C (sec)	D (%)	E (sec)	F (sec)	G (%)
1	-	-	-	-	-	-
2	-	-	-	-	-	-
3	196	372	20	202	378	15
4	170	358	35	175	363	30
5	154	339	40	159	341	40
6	139	326	55	144	334	55
7	120	319	75	129	330	70
8	109	309	80	113	314	80
9	99	298	85	104	304	80
10	91	283	100	95	293	100

Table 13. Perception and feeding response stimulation of glycogen. *see* legend explanation in text to Table 1.

A (%)	B (sec)	C (sec)	D (%)	E (sec)	F (sec)	G (%)
1	70	178	75	79	181	70
2	60	162	90	67	163	85
3	47	152	100	55	158	100
4	40	144	100	47	149	100
5	32	136	100	39	141	100
6	27	127	100	32	129	100
7	22	114	100	26	119	100
8	16	103	100	19	109	100
9	13	95	100	16	102	100
10	10	88	100	13	92	100

Table 14. Response (%) realtive to the concentration (%) of salts. A: Concentration, B: Potassium chloride (KCl), C: Potassium bromide (KBr), D: Potassium iodide (KI), E:Sodium nitrate (Na NO_3), F: Ammonium chloride (NH_4Cl), G: Lithium chloride (LiCl), H: Magnesium chloride ($MgCl_2$), I: Sodium chloride (NaCl), J: Calcium chloride ($CaCl_2$), K: Copper sulphate ($CuSo_4$). P - Positive response; ED = Escape or Defensive response.

B. zeylonica

A %	B ED	C ED	D ED	E ED	F ED	G P	H P	I P	J ED	K ED
1	-	-	70	80	65	65	35	-	-	-
2	-	-	80	85	80	70	50	-	-	-
3	10	-	90	100	100	75	60	-	-	-
4	15	-	100	100	100	80	75	-	10	5
5	35	-	100	100	100	100	80	-	30	20
6	45	5	100	100	100	100	100	-	40	30
7	60	15	100	100	100	100	100	-	55	55
8	75	30	100	100	100	100	100	-	70	60
9	90	40	100	100	100	100	100	-	80	75
10	100	55	100	100	100	100	100	10	100	80
11	100	70	100	100	100	100	100	10	100	100
12	100	80	100	100	100	100	100	20	100	100
13	100	85	100	100	100	100	100	40	100	100
14	100	90	100	100	100	100	100	50	100	100
15	100	100	100	100	100	100	100	65	100	100
16	100	100	100	100	100	100	100	65	100	100
17	100	100	100	100	100	100	100	80	100	100
18	100	100	100	100	100	100	100	100	100	100
19	100	100	100	100	100	100	100	100	100	100
20	100	100	100	100	100	100	100	100	100	100

B. spirata

A %	B ED	C ED	D ED	E ED	F ED	G P	H P	I P	J ED	K ED
1	-	-	55	60	50	10	-	-	-	-
2	-	-	60	70	60	25	-	-	-	-
3	-	-	70	85	75	30	20	-	-	-
4	10	-	85	85	90	50	40	-	-	-
5	20	-	100	90	100	65	50	-	10	10
6	25	-	100	100	100	70	65	-	25	20
7	30	10	100	100	100	80	70	-	40	40
8	40	15	100	100	100	100	80	-	60	55
9	55	25	100	100	100	100	100	-	70	70
10	70	30	100	100	100	100	100	-	75	85
11	80	40	100	100	100	100	100	-	80	90
12	80	40	100	100	100	100	100	10	100	100
13	100	55	100	100	100	100	100	20	100	100
14	100	70	100	100	100	100	100	30	100	100
15	100	80	100	100	100	100	100	55	100	100
16	100	90	100	100	100	100	100	70	100	100
17	100	100	100	100	100	100	100	70	100	100
18	100	100	100	100	100	100	100	90	100	100
19	100	100	100	100	100	100	100	100	100	100
20	100	100	100	100	100	100	100	100	100	100

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