# New Insect Juvenile Hormone Mimics: Aromatic Schiff Bases and Related Compounds Against the Large Milkweed Bug and Yellow Mealworm

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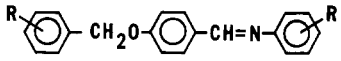
Schiff bases derived from substituted 4-(benzyloxy)benzaldehyde and 2,6-difluoroaniline were highly effective nonterpenoid juvenile hormone (JH) mimics for *Oncopeltus fasciatus* (Dallas) when applied topically to fifth instar nymphs. *N*-[4-(Benzyloxy)benzyl]anilines, obtained by reduction of Schiff bases, were more effective than the corresponding Schiff bases in *Oncopeltus* tests. *N*-[[4-[(3-Chlorophenyl)methoxy]phenyl]methyl]-2,6-difluorobenzenamine (**39**) was the most effective JH mimic of the compounds tested. Although oviposition was normal, egg hatch was severely inhibited when adult *Oncopeltus* of both sexes were topically treated with this compound or with 2,6-difluoro-*N*-[[4-(phenylmethoxy)phenyl]methylene]benzenamine (**23**). The Schiff bases and their reduction products were less effective against *Tenebrio molitor* L. than against *Oncopeltus*.

Most of the earlier and indeed many of the more recently developed juvenile hormone (JH) analogues have terpenoid or sesquiterpenoid structure (Wigglesworth, 1970; Bowers, 1971; Menn and Beroza, 1972; Sláma et al., 1974). However, several classes of nonterpenoid JH mimics have also been reported (Bowers, 1968; Zaoral and Sláma, 1970; Punja et al., 1973; Sláma et al., 1974; Franke et al., 1975a–d; Pallos et al., 1976; Nilles et al., 1977), but the total number of highly effective compounds in these classes is comparatively low. Since terpenoid JH mimics are often costly to synthesize or are unstable under field conditions, pesticide chemists are constantly searching for new structures to minimize or alleviate these problems.

In our screening program, the Schiff base 2-fluoro-*N*-[[4-(phenylmethoxy)phenyl]methylene]benzenamine (**2**; Table I) was effective as a JH mimic for the large milkweed bug, *Oncopeltus fasciatus* (Dallas), when administered topically. Although **2** was only moderately effective in our tests, the novelty of this nonterpenoid JH mimic prompted us to further investigate this class of compounds. We report here the JH effectiveness of 36 Schiff bases (structure A) and four related derivatives when they were



administered topically to immature large milkweed bugs and yellow mealworms, *Tenebrio molitor* L.

## MATERIALS AND METHODS

**Chemicals.** Melting points are uncorrected. Microanalyses were performed by Galbraith Laboratories, Inc., Knoxville, TN. All synthesized compounds gave acceptable (±0.4%) analyses for C, H, and N. Tables of complete analytical data for compounds will appear in the microfilm edition (see paragraph at end of paper regarding supplementary materials).

Most of the compounds listed in Table I were purchased from Aldrich Chemical Co., Inc., Alfred Bader Chemicals Div., Milwaukee, WI, and were used without further purification. Compounds **3**, **23**, **24**, and **25** (Table I) were synthesized and had melting points of 77–79.5, 88–90, 81–83, and 84.5–87 °C, respectively. Compound **2** was also synthesized and its melting point, infrared spectrum, and $R_f$ value (TLC) were identical with that of the purchased sample. Bioassay results of synthesized **2** were identical with those obtained with the purchased sample. All compounds listed in Table II were synthesized.

Schiff bases were prepared by the following procedure.

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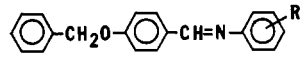
Table I. JH Mimicking Activity for Schiff Bases Applied Topically to *Oncopeltus* Nymphs



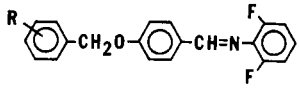
| compd no. | R | av JH rating at 10 μg/insect[a] |
|---|---|---|
| 1 | H | 1.5[c] |
| 2 | 2-F | 2.0 |
| 3 | 2-Cl | 1.9 |
| 4 | 2-$CF_3$ | *d* |
| 5 | 3-Br | *e* |
| 6 | 3-$CF_3$ | *e* |
| 7 | 3-$NO_2$ | 1.0[b] |
| 8 | 4-F | 2.5[b] |
| 9 | 4-Cl | 0.0 |
| 10 | 4-Br | *e* |
| 11 | 4-I | *e* |
| 12 | 4-$CH_3$ | *e* |
| 13 | 4-$C_2H_5$ | 2.0[c] |
| 14 | 4-$CH_3O$ | *e* |
| 15 | 4-$CO_2H$ | 0.0 |
| 16 | 4-$CO_2C_2H_5$ | *e* |
| 17 | 3,4-$(CH_3)_2$ | 0.3 |
| 18 | 3,4-$Cl_2$ | 0.0 |
| 19 | 2-$CH_3$, 5-$NO_2$ | *e* |
| 20 | 3-$NO_2$, 4-$CH_3$ | *d* |
| 21 | 2-Br, 4-$CH_3$ | *e* |
| 22 | 2,4-$(CH_3O)_2$, 5-Cl | *d* |
| 23 | 2,6-$F_2$ | 3.0 |
| 24 | 2,6-$Cl_2$ | 0.6 |
| 25 | 2,6-$Br_2$ | *e* |

[a] *Oncopeltus* JH ratings: 0 = normal adult; 1 = normal adult except for retention of nymphal coloration on the abdomen; 2 = adult with smaller wings and nymphal coloration on the abdomen; 3 = supernumerary nymph. Sesamex had a 3.0 rating at 10 μg and 0.2 rating at 1.0 μg. Acetone check had a 0.0 rating. [b] Forty percent died attempting to molt. [c] Sixty percent died attempting to molt. [d] All died attempting to molt. [e] Rated 0.0 at maximum test concentration (50 μg).

A mixture of 4-(benzyloxy or substituted 4-benzyloxy)-benzaldehyde (0.02 mol), the appropriately substituted aniline (0.02 mol), *p*-toluenesulfonic acid (25–50 mg), and benzene (100 mL) was heated under reflux for ca. 3–6 h, or until all the water (0.02 mol) was removed by azeotropic distillation. The solvent was removed on a rotary evaporator and the residue was recrystallized from the appropriate solvent (yields after recrystallization, 30–70%). Based on previous information (De Gaouck and LeFèvre, 1938; Layer, 1963; McCarty, 1970), the benzyloxyphenyl and aniline portions of the molecule are considered to have a trans configuration.

The substituted 4-(benzyloxy)benzaldehydes were synthesized by the following procedure. An ethanolic

Table II. JH Mimicking Activity for Schiff Bases Applied Topically to Immature *Oncopeltus* and *Tenebrio*



| compd no. | R | mp, °C[a] | av JH ratings at indicated dose (μg/insect)[b] *Oncopeltus* 10 | 1.0 | 0.1 | 0.01 | *Tenebrio* 50 |
|---|---|---|---|---|---|---|---|
| **23** | H | 88-90 | 3.0 | 3.0 | 1.2 | 0.0 | 1.6 |
| **26** | 2-Cl | 99-101.5 (54-56) | *f* | | | | 0.0 |
| **27** | 3-Cl | 105.5-108 (53-55) | 3.0 | 3.0 | 3.0 | 2.0[c] | 0.0 |
| **28** | 4-Cl | 115-117 (73-74.5) | 3.0[c] | 3.0[c] | *e* | 0.0 | 0.4 |
| **29** | 2-F | 73-74 (57-60) | 3.0 | 2.0[d] | 0.0 | | 1.8 |
| **30** | 3-F | 74.5-76.5 (43-45) | 3.0[c] | 3.0 | 3.0 | 2.0 | 0.2 |
| **31** | 4-F | 96-97 (98-100) | 3.0 | 3.0 | 3.0[d] | 0.0 | 0.4 |
| **32** | 2-$CF_3$ | 48-50.5 (120/0.2 mm) | *f* | | | | 0.0 |
| **33** | 3-$CF_3$ | 83-84 (46-48) | *f* | | | | 0.0 |
| **34** | 4-$CF_3$ | 128-129 (112/0.13 mm) | *f* | | | | 0.0 |
| **35** | 2,4-$Cl_2$ | 109-110 (98-99) | 1.6 | 1.0 | 0.0 | | 0.2 |
| **36** | 3,4-$Cl_2$ | 105-107 (91-93) | 3.0 | *e* | *e* | 0.4 | 0.0 |

[a] Values in parentheses are melting points or boiling points for 4-(benzyloxy)benzaldehyde precursors. [b] See Table I footnotes for key to *Oncopeltus* JH ratings. *Tenebrio* JH ratings: 0 = normal adult; 1 = retention of gin traps or urogomphi; 2 = retention of both gin traps and urogomphi; 3 = intermediate, retention of both gin traps and urogomphi plus retention of pupal cuticle around treated area, e.g., the whitish neutral abdominal coloration; 4 = second pupae, retention of all pupal characteristics. Farnesyl methyl ether rated 4.0 at 10 μg. [c] Sixty percent died attempting to molt. [d] Eighty percent died attempting to molt. [e] One-hundred percent died attempting to molt. [f] No test run at 10 μg. Rated 0.0-1.0 at 50 μg.

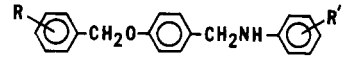
solution containing 0.1 mol of the sodium salt of 4-hydroxybenzaldehyde [from NaOH (0.1 mol), 4-hydroxybenzaldehyde (0.1 mol), and EtOH (100 mL)] and 0.1 mol of the appropriately substituted benzyl chloride (or bromide) was heated under reflux (argon atmosphere) for 3–12 h. The hot mixture was filtered to remove insoluble salts and then cooled to crystallize the product. Highly soluble products were obtained by removing the ethanol and extracting the residue with diethyl ether or methylene chloride (yields after recrystallization, 50–83%). Melting points for the (benzyloxy)benzaldehydes are reported in Table II.

The *N*-[4-(benzyloxy or substituted 4-benzyloxy)-benzyl]anilines **37–40** (Table III) were prepared by sodium borohydride reduction of the corresponding Schiff bases by using a slightly modified procedure (7:1 w/w ratio of $NaBH_4$ to Schiff base; reaction time, ca. 3–4 h at 15 °C, then 0.75 h at 60 °C) of Billman and Diesing (1957). Reduction products were purified by column chromatography (alumina) and, except for **37** (mp 60–62 °C), were viscous oils (yields, 50–75%). Proton magnetic resonance spectra confirmed the identities of reduced products. The purities of reduced products were estimated by TLC to be greater than 95%.

**Biological Tests.** Details of the JH-activity test used with large milkweed bugs were described by Redfern et al. (1971). Briefly, compounds (purified by recrystallization or chromatography) were dissolved in acetone and the desired dose was applied topically (1 μL/nymph) with a calibrated glass micropipet to the last abdominal segments of the ventral side of 2- to 8-h-old fifth instars. Treated nymphs were held until they molted to adults or until some morphological changes characteristic of juvenilization were observed. Rating systems used for JH effects listed in Table I footnotes.

The JH-activity tests used with yellow mealworms were performed by the method of Redfern et al. (1970) except that a disposable, calibrated glass micropipet was used to administer the compounds. Briefly, acetone solutions of the compounds were topically applied (1 μL/pupa delivering 50 μg of test compound) to the terminal abdominal segments of the ventral side of 2- to 8-h-old pupae (10 pupae/test). The pupae were held until they molted or until JH morphological effects were noted. Rating system used for JH effects listed in Table II footnotes.

Table III. JH Mimicking Activity for Reduced Schiff Bases Applied Topically to Immature *Oncopeltus* and *Tenebrio*



| compd no. | R | R′ | av JH rating at indicated dose (μg/insect)[a,b] *Oncopeltus* 1.0 | 0.1 | 0.01 | 0.001 | *Tenebrio* 50 |
|---|---|---|---|---|---|---|---|
| **37** | H | 2-F | 0.2 | 0.0 | | | 1.6 |
| **38** | H | 2,6-$F_2$ | 3.0 | 3.0 | 0.0 | | 2.0 |
| **39** | 3-Cl | 2,6-$F_2$ | 3.0 | 3.0 | 3.0 | 2.5[c] | 0.0 |
| **40** | 3-F | 2,6-$F_2$ | 3.0 | 3.0 | 3.0 | 0.0 | 1.2 |

[a] See footnotes Tables I and II for key to JH ratings. [b] All reduction products rated 3.0 at 10 μg. [c] Sixty percent died attempting to molt.

## RESULTS AND DISCUSSION

Structure–activity relationships of Schiff bases were derived from three series of compounds. Analogues in the first series, **1–25** (Table I), retained the basic skeletal structure of **2**, and modifications were made in the aniline portion of the molecule. Analogues in the second series, **26–36** (Table II), retained the most effective aniline moiety (determined from the first series), and modifications were made in the aldehydic portion of the molecule by introducing various substituents on the terminal benzene ring. In the final series, highly effective Schiff bases identified from the previous series were reduced to the corresponding *N*-[4-(benzyloxy)benzyl]anilines, **37–40** (Table III).

Of the 24 analogues of **2** (Table I) tested against *Oncopeltus*, the 2,6-difluoro analogue, **23**, was the most effective. Replacement of fluorine in **23** with chlorine (**24**) or with bromine (**25**) caused either substantial or total loss of JH activity. Schiff bases **3** (2-Cl), **8** (4-F), and **13** (4-$C_2H_5$) were as effective as **2**, but **8** and **13** were highly toxic to nymphs. JH scores could not be assessed for **4**,

20, and 22 since all insects died at the concentration tested.

Since the 2,6-difluoroaniline moiety gave optimum results, additional analogues 26–36 (Table II) containing this group were synthesized. Three analogues, 27, 30, and 31, bearing halogen substituents on the terminal benzene ring were more effective than unsubstituted 23 in *Oncopeltus* tests. Compounds 28 and 36 were also effective, but toxic effects at low concentrations prevented adequate assessment of JH activity.

The location of substituents appeared to be an important criterion for activity. For example, test results in *Oncopeltus* for the two isomeric series (R = Cl or F) showed activity to parallel the following order of substitution: meta > para > ortho. The increased effectiveness of the disubstituted analogue 36 (R = 3,4-$Cl_2$) over its isomer 35 (R = 2,4-$Cl_2$) supports these positional requirements. Surprisingly, the highly lipophilic trifluoromethyl analogues (32–34) were either slightly effective or totally ineffective, suggesting the possibility of a "size" requirement for meta substituents.

Reduction of highly effective Schiff bases 2, 23, 27, and 30 gave secondary amines 37–40 (Table III). Except for 37, amines 38–40 were more effective against *Oncopeltus* than the corresponding Schiff bases. Of all compounds tested against *Oncopeltus*, 39 was the most effective JH mimic, eliciting a JH score of 2.5 at a dose of 1 ng/insect.

Schiff bases and their reduced products were much less effective against *Tenebrio*. Although Schiff bases 23 and 29 (Table II) and amines 37, 38, and 40 (Table III) elicited modest JH effects in *Tenebrio*, the dose required to produce these effects was high (50 μg/pupa). Interestingly, compound 39, the most effective in *Oncopeltus*, was totally ineffective in *Tenebrio*.

Results from sterility tests with 23 and 39 showed that reproduction of *Oncopeltus* was also affected. For example, pairs consisting of an adult of either sex that had been treated topically with 23 (100 μg/insect) or 39 (10 μg/insect) showed normal oviposition, but 98–100% of resultant eggs failed to hatch. Neither 23 nor 39 was exceedingly toxic to adults in topical treatments since mortality was only 10% at 5 days posttreatment for both compounds at the 100 μg/insect dose.

In summary, we have identified 11 new, easily synthesized, and highly effective, nonterpenoid JH mimics of *Oncopeltus*, and we have identified some important structure–activity relationships for this class of compounds. Although Schiff bases themselves may present potential problems in hydrolytic stability, their corresponding reduction products are less likely to do so. Also, the amines, because of their high aromatic content ought to provide greater overall compound stability (i.e., photolytic, oxidative, thermolytic, etc.) than the classical, highly unsaturated, aliphatic juvenoids currently in use or under investigation. Finally, since preliminary data from our laboratories indicate that other insects can be similarly affected by derivatives related to those described in the present study, we speculate that a variety of potentially useful JH mimics bearing the benzyloxyphenyl moiety may be developed.

ACKNOWLEDGMENT

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**Supplementary Material Available:** A listing of analytical data and recrystallization solvents for the analogues (1 page). Ordering information is given on any current masthead page.

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