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P_2O_5/SiO_2-Catalyzed solvent-free room-temperature synthesis of 2-aryl-benzimidazoles and their toxicity against *Artemia salina*

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Abstract

Twelve (12) 2-Aryl-1*H*-benzimidazole derivatives were synthesized from a series of 4-*o*-phenylenediamines [R: -NO_2, -$COOCH_2CH_3$, and -COAr] and 4-aryl aldehydes [R: -H, -$N(CH_2CH_3)_2$, -$CH(CH_3)_2$, -$CH(OCH_2CH_3)_2$] utilizing a simple, cheap, and heterogeneous P_2O_5/SiO_2 catalytic system at room temperature and solvent-free conditions. Moreover, the bioactivity of the prepared benzimidazoles, evaluated *via* brine shrimp lethality assay (BSLA), showed that almost all benzimidazole products were cytotoxic, except for 5-nitro-2-phenyl-1*H*-benzo[d]imidazole (5a). Highest activity was observed for 2-(4-isoproylphenyl)-5-nitro-1*H*-benzo[d]imidazole (5b) with acute lethal concentration (LC_{50}) of 43.42 ppm, whereas the least active *N, N*-diethyl-4-(5-nitro-1*H*-benzo[d]imidazol-2-yl) aniline (5c), had chronic LC_{50} value of 283.39 ppm. Qualitative structure - BSLA activity correlation showed that balanced electron-donating capacity of R_2 coupled with electron-withdrawing effect of R_1 led to enhanced toxicity of 2-Aryl-1*H*-benzimidazoles.

Keywords: benzimidazoles; green chemistry; P_2O_5/SiO_2; room-temperature synthesis

1. Introduction

Benzimidazoles are a class of compounds that are relatively abundant in nature but can also be chemically synthesized through a variety of synthetic routes. The resemblance and occurrence of their structure to many natural biomolecules gave rise to their emphasized broad spectra of biological activities and therapeutic effects [1]. This includes being antiviral, antimicrobial, antiparasitic, antihypertensive, anti-inflammatory, anti-ulcer, and anticancer among others [2–4]. Furthermore, non-pharmaceutical applications have also emerged for these molecules in the manufacture of textile industry formulations, as well as in the synthesis of azo and fluorescent dyes [5]. They are also considered as important novel components in organic light emitting diodes (OLED's) [6], optoelectronic switches [7], and fuel cells [8] in the electronics industry.

The countless benefits of benzimidazole derivatives, especially in the medical field, have resulted to a drive to further develop efficient, clean, and more economically feasible methods for their synthesis. 2-Arylbenzimidazoles are among the type of benzimidazoles that have been extensively studied as demonstrated by a large number of scientific literatures published concerning their synthetic process [1]. Synthesis of 2-Arylbenzimidazole from oxidative coupling of *o*-phenylenediamine and aryl aldehydes is the most popular route due the availability of a vast number of aldehydes, mild to moderate reactions conditions, and much faster reaction times. Thus several 2-Arylbenzimidazoles using this preferred route were made with conventional protocols involve the use of strong mineral acids, such as HCl, or oxidizing agents like $K_3Fe(CN)_6$ under basic condition, $Mn(OAc)_3$ in AcOH, CAN-H_2O_2, $Cu_{3/2}PMo_{12}O_{40}/SiO_2$, and $Fe(NO_3)_3$ - H_2O_2 [1,9]. Although majority of the methods are effective, many suffer from one or more disadvantages such as (a) poor yields, (b) use of expensive reagents, (c) multistage synthetic approaches, (d) tedious workup procedures, and (e) occurrence of several side reactions [10]. Because of this, introduction of new methods and/or further work on technical improvements of procedures to overcome these limitations is still an important experimental challenge.

With the advent of green chemistry, new techniques have been designed which not only are more efficient but are also lacking in the drawbacks posed by more traditional approaches. Such methods make use of milder reaction conditions, cheaper and more readily available starting materials, minimal use of organic solvents, and utilization of less hazardous chemicals [11]. Currently, the solvent-free synthetic procedure paired with a inexpensive recyclable heterogeneous catalyst being developed are encouraged to be more inclined to green chemistry principles. The use of P_2O_5/SiO_2 catalyst had the advantage in handling and storage, highly reactive, and clean reaction that can be worked up by simple filtration. Moreover, it can be easily modified and fine-tuned as compared to traditional synthetic methods.

The brine shrimp lethality assay (BSLA) that uses *Artemia salina* nauplii is a versatile and cost-effective bioassay that can effectively identify bioactivity in organic compounds [12] . BSLA serves as an efficient tool for assessing general toxicity detection and indicating biological activity. Its affordability, simplicity in execution, and ready availability of inexpensive brine shrimp eggs contribute to making BSLA an invaluable method.

In this study, 2-Aryl-1*H*-benzimidazole derivatives were synthesized *via* a solvent-free reaction, using P_2O_5/SiO_2 as reusable heterogeneous catalyst under ambient conditions. The potential of the synthesized benzimidazole derivatives as active pharmaceutical ingredients were assessed by evaluating their toxicity toward brine shrimps.

2. Materials and methods

2.1 Starting Materials and Reagents

The starting materials, reagents, and catalysts used in this study were purchased from commercial suppliers and utilized as received.

2.2 Preparation of Phosphorus Pentoxide Supported by Silica gel (P_2O_5/SiO_2) Catalyst

The catalyst mixture was prepared by mixing 0.80g of phosphorus pentoxide (P_2O_5) with 1.20g of Wakogel C200, a chromatography-grade silica gel (SiO_2), in a mortar and pestle with vigorous grinding to give around 2.00g white powder catalyst system [13,14].

2.3 Catalyst Optimization

The ideal amount of catalyst for the proposed solvent-free room-temperature synthesis was determined through three model reactions which made use of three o-phenylenediamine analogs paired with benzaldehyde in a 1:2 mol ratio. Different quantities (0.01g, 0.02g, 0.03g, 0.04g, 0.05g) of the prepared (P_2O_5/SiO_2) catalyst were used for the model reaction. The amount of catalyst that required the shortest time for the reaction to come to completion, as monitored by thin layer chromatography (TLC) was used as the standard amount for the succeeding synthesis of the remaining benzimidazole derivatives.

2.4 Solvent-free Room-temperature Synthesis of 2-Aryl-1H-benzimidazoles

The selected o-phenylenediamine (3 mmol) and benzaldehyde (6 mmol) congeners were mixed in a 15-mL test tube together with 0.02 g of the P_2O_5/SiO_2 catalyst and subjected to vortex mixing at room temperature. The progress of the reaction was monitored using TLC with hexane: ethyl acetate (1:4) as the solvent system. Upon completion of the reaction, the resulting mixture was added with 10 mL of ethanol and subjected to centrifugation to separate the catalyst from the mixture. The liquid ethanolic portion was afterwards collected and the process was repeated 3 times. Subsequently, the collected ethanolic portions were pooled and evaporated under reduced pressure to afford the target benzimidazoles. The crude product was recrystallized in ethyl acetate, collected and dried.

2.5 Brine Shrimp Lethality Assay

Procedure for the BSLA experiment was based on the protocol by Ngutaa and Mbaria [15] with slight modifications. Stock solutions of 50,000 ppm of the synthesized 2-Aryl-1*H*-benzimidazoles were prepared by dissolving 50 mg of each benzimidazole sample with 100 µL of dimethyl sulfoxide DMSO followed by dilution to 1000 µL with filtered, sterilized seawater. Three solutions with different concentrations of each benzimidazole sample (10, 100, 1000 ppm) and three replicates each were prepared from the stock solutions. Each solution contained 10 nauplii delivered from the incubation vessel and had a final volume of 5 mL. The test tubes were left uncovered in a well-ventilated place under constant illumination. The number of surviving brine shrimp nauplii were counted and recorded after 6 and 24 hours and the percent deaths for each dose was calculated. In

order to make certain that the mortality observed in the bioassay is only caused by the biological activity of 2-Aryl-1*H*-benzimidazoles and not of starvation or associated toxicity of DMSO, controls were also run parallel to the test samples. The total number of dead and surviving shrimp nauplii for each corresponding time (6 hours and 24 hours) were employed for the calculation and estimation of the lethal concentration (LC_{50}) value using Miller and Tainter method (modified probit analysis) [16–19].

3. Results and discussion

3.1 Optimization of P_2O_5/SiO_2 catalyst

The catalyst system consisting of phosphorus pentoxide (P_2O_5) supported in silica gel (SiO_2) was prepared by simple grinding process using mortar and pestle. To determine the ideal amount of the catalyst to be used for the synthesis of the target compounds, three model reactions were used (Figure 1).

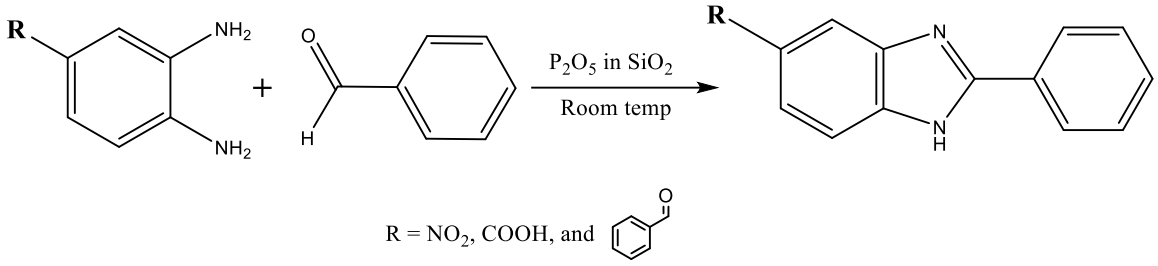


Figure 1 Model reaction for the optimization of P_2O_5/SiO_2 catalyst system.

A concentration of 8.0% w/w P_2O_5 in SiO_2 was initially used for this optimization. For each model reaction, different amounts of the catalyst were utilized. The catalyst amount that gave the shortest reaction completion time, as assessed by the disappearance of starting materials in TLC profiles, was regarded as the ideal amount and was utilized for the synthesis of 2-aryl-1*H*-benzimidazoles. Figure 2 shows the summary of the results for the initial test using 8.0% w/w P_2O_5 in SiO_2.

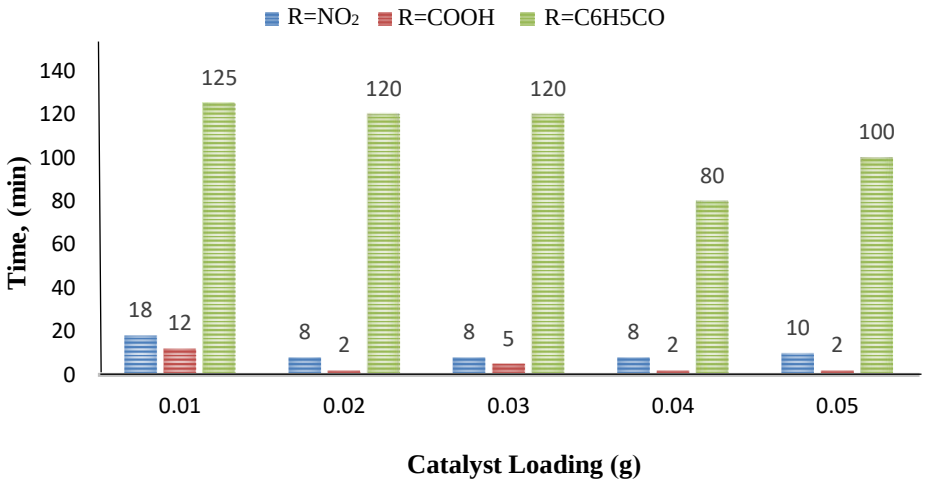


Figure 2 Required reaction completion time for different catalyst loading of 8% P_2O_5/SiO_2 catalyst system.

It was observed that P_2O_5 interacts readily with moisture upon exposure to open environment. However, by supporting it in silica gel, the stability of P_2O_5 in ambient conditions as well as its ease of handling was significantly increased. Initial results indicated that the reaction of 4-nitro-*o*-phenylenediamine and benzaldehyde was completed at 8.0 min and 0.02 g of catalyst. On the other hand, the reaction of benzaldehyde with 3,4-diaminobenzoic acid was completed after 2.0 min and 0.02 g of catalyst. Meanwhile, much longer times were required for the reaction of benzaldehyde and 3,4-diaminobenzophenone with the shortest reaction time of 80 min at 0.04 g catalyst. This reaction behavior can be attributed to the bulkiness of the benzophenone group which may have induced steric effects leading to much longer reaction times. Aside from those observations, it was also

noticed that for the three baseline reactions prepared, increasing the amount of catalyst also increased the complexity of the mixing process. This intricacy is highly pronounced for all reactions as the amount of catalyst is increased from 0.03 g to 0.05 g. Inefficient mixing had led to much unreacted starting material thus considered to be unproductive, wasteful, and uneconomical.

Based on the results for the model reactions, the best amount of the catalyst to be used for the synthesis of 2-Aryl-1*H*-benzimidazole was 0.02 g. In spite of this, the problem of longer reaction times for 3,4-diaminobenzophenone needed to be addressed. Thus, there was an initiative to increase the concentration of the catalyst system from 8.0% w/w to 40.0% w/w P_2O_5 in SiO_2 parallel to the reported catalyst concentration used by Eshghi's group of 37% w/w P_2O_5 in SiO_2 in different organic reactions [20]. From the predetermined optimum catalyst amount of 0.02 g, the catalyst concentration was increased to 40% w/w (Figure 3).

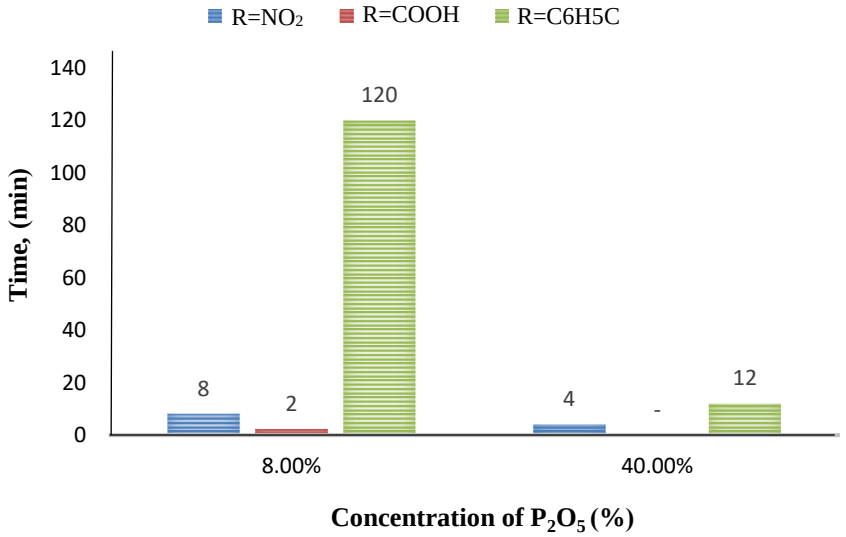


Figure 3 Required reaction completion time for model reactions at different concentrations of P_2O_5 in the P_2O_5/SiO_2 catalytic system.

As demonstrated above, improved reaction times were achieved most especially on the reaction using 3,4-diaminobenzophenone as starting material. However, for the reaction of 3,4-diaminobenzoic acid with benzaldehyde, it was observed that as the catalyst concentration was increased, numerous side products were formed as evidenced by its TLC profile. The reaction behavior of 3,4-diaminobenzoic acid toward the significant production of side products may be attributed to the presence of carboxylic acid functional group that was potentially activated by the increased P_2O_5 concentration even under ambient and mild conditions. This activation may have led the carboxylic acid group to react with the amino groups present on 3,4-diaminobenzoic acid, leading to the formation of side products.

To minimize the problems encountered with 3,4-diaminobenzoic acid, the carboxylic acid group was esterified with ethanol. The conversion of carboxylic to ester would reduce the formation of side products since ester group is much less reactive toward amines in the presence of P_2O_5.

3.2 Solvent-free Room-Temperature Synthesis of 2-aryl-1H-benzimidazoles

The synthesis of the desired 2-Aryl-1*H*-benzimidazoles (Figure 4) was done using the protocol described in the methods section and the identified ideal concentration and amount of catalyst.

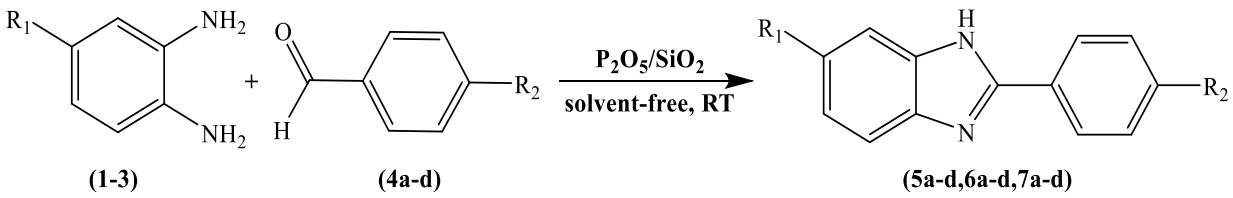


Figure 4 General reaction for the synthesis of 2-aryl-1*H*-benzimidazoles.

Table 1 Summary of product yields for synthesized 2-aryl-1*H*-benzimidazoles.

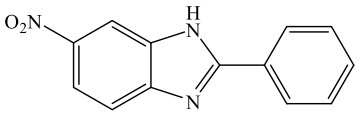
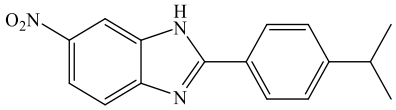
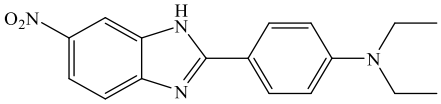
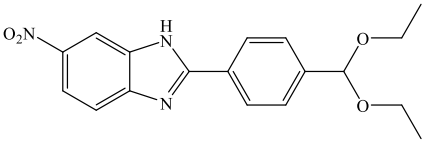
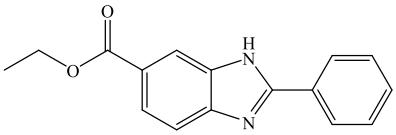
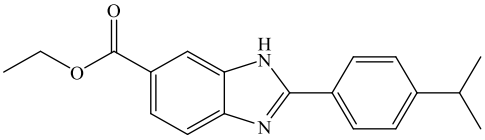
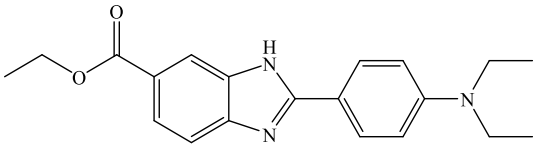
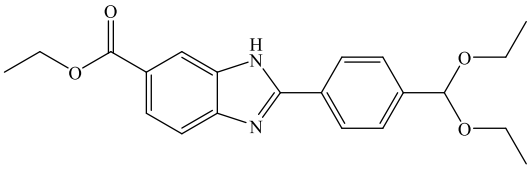
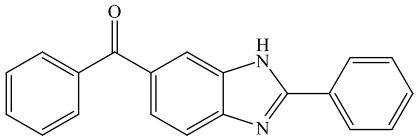
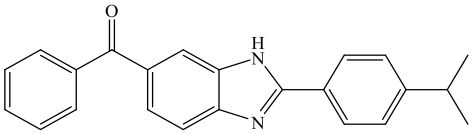
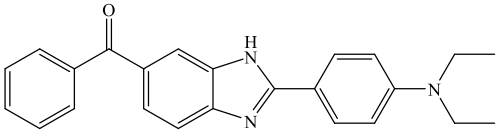
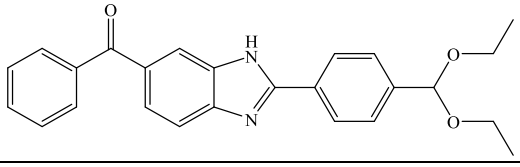
Product	Structure	Reaction Time (min)	Yield (%)
5a		4	16.5
5b		5	15.1
5c		60	13.7
5d		25	18.9
6a		10	10.9
6b		10	10.6
6c		30	5.0
6d		15	9.9
7a		12	19.2
7b		12	14.9
7c		30	12.8
7d		15	11.8

Table 2 Melting point, IR, and ^{1}H-NMR data for synthesized 2-aryl-1*H*-benzimidazoles.

Product	Morphology	Melting point (°C)	IR (ATR, cm^{-1})	^{1}H-NMR (500 MHz, DMSO-d_6)
5a	White powder	>220	ν 1633.37 (C=N), 1094.55 (C-N)	8.46 (s, 1H, N-H), 8.21 (d, 2H, Ar-H), 8.21 (d, 1H, Ar-H), 8.11 (1H-Ar-H), 7.75 (d, 1H, Ar-H), 7.57 - 7.60 (m, 3H, Ar-H),
5b	White powder	>220	ν 2969.08 (N-H), 1633.01 (C=N), 1191.24 (C-N)	8.44 (s, 1H, N-H), 8.13 (d, 2H, Ar-H), 8.13 (d, 1H-Ar-H), 8.10 (dd, 1H, Ar-H), 7.73 (d, 1H, Ar-H), 7.45 (d, 2H, Ar-H), 2.92 - 3.00 (m, 1H, CH), 2.23 (d, 6H, CH_3),
5c	Yellow powder	>220	ν 2974.96 (N-H), 1601.36 (C=N), 1194.65 (C-N)	8.34 (s, 1H, N-H), 8.06 (d, 1H, Ar-H), 8.13 (d, 3H, Ar-H), 7.63 (d, 1H, Ar-H), 6.77 (d, 2H, Ar-H), 3.39 (q, 4H, CH_2), 1.10 (t, 6H, CH_3),
5d	Yellow powder	>220	ν 2976.39 (N-H), 1632.83 (C=N), 1215.23 (C-N)	10.08 (s, 1H, N-H), 8.39 (d, 2H, Ar-H), 8.14 (1H, Ar-H), 8.12 (1H, Ar-H), 7.75-7.82 (m, 3H, Ar-H), 5.56(s, 1H, CH), 3.43 (q, 4H, CH_2), 1.04 (t, 6H, CH_3),
6a	Brown powder	>220	ν 1625.12 (C=N), 1214.98 (C-N)	7.84 (s, 1H, N-H), 8.18-8.20 (m, 3H, Ar-H), 7.68 (d, 1H, Ar-H), 7.51 - 7.69 (m, 4H, Ar-H), 4.32 (q, 2H, CH_2), 1.34 (t, 3H, CH_3)
6b	Brown powder	>220	ν 2963.07 (N-H), 1625.38 (C=N), 1212.38 (C-N)	8.19 (s, 1H, N-H), 8.12 (d, 2H, Ar-H), 7.84 (d, 1H-Ar-H), 7.66 (d, 1H, Ar-H), 77.57 - 7.60 (m, 3H, Ar-H), 4.33 (q, 2H, CH_2), 2.93 - 2.98 (m, 1H, CH), 1.34 (t, 3H, CH_3), 1.24 (d, 6H, CH_3)
6c	Brown powder	>220	ν 2972.18 (N-H), 1603.73 (C=N), 1207.54 (C-N)	8.10 (s, 1H, N-H), 7.94 - 8.02 (m, 3H, Ar-H), 7.78 (d, 1H, Ar-H), 7.57 (d, 1H, Ar-H), 6.78 (d, 2H, Ar-H), 4.31 (q, 2H, CH_2), 3.40 (q, 4H, CH_2), 1.34 (t, 3H, CH_3), 1.11 (t, 6H, CH_3)
6d	Brown powder	>220	ν 2976.37 (N-H), 1626.29 (C=N), 1211.75 (C-N)	10.07 (s, 1H, N-H), 8.39 (d, 2H, Ar-H), 8.19 (d, 1H, Ar-H), 8.08 (d, 2H-Ar-H), 7.87 (d, 1H, Ar-H), 7.71 (d, 1H, Ar-H), 5.55 (1H, CH), 4.32 (q, 4H, CH_2), 3.43 (q, 2H, CH_2), 1.34 (t, 6H, CH_3), 1.04 (t, 3H, CH_3)
7a	White powder	>220	ν 1651.80 (C=N), 1224.86 (C-N)	7.97 (s, 1H, N-H), 8.21 (d, 2H, Ar-H), 7.74 - 7.77 (m, 4H, Ar-H), 7.65 - 7.70 (m, 3H, Ar-H), 7.57 - 7.60 (m, 4H, Ar-H)
7b	White powder	>220	ν 2963.41 (N-H), 1656.90 (C=N), 1221.59 (C-N)	7.93 (s, 1H, N-H), 8.13 (d, 2H, Ar-H), 7.76 (d, 2H, Ar-H), 7.72 - 7.73 (m, 1H, Ar-H), 7.68 (d, 2H, Ar-H), 7.57 (d, 2H, Ar-H), 7.56 (1H, Ar-H), 7.44 (d, 2H, Ar-H), 2.92 - 3.00 (m, 1H, CH), 1.24 (d, 6H, CH_3)
7c	Light yellow powder	>220	ν 2969.64 (N-H), 1654.97 (C=N), 1220.76 (C-N);	7.86 (s, 1H, N-H), 7.95 (d, 2H, Ar-H), 7.72 (d, 2H, Ar-H), 7.63 - 7.67 (m, 2H, Ar-H), 7.59 - 7.64 (m, 2H, Ar-H), 7.57 (1H, Ar-H), 7.54 - 7.57 (m, 1H, Ar-H), 6.77 (d, 2H, Ar-H), 3.39 (q, 4H, CH_2), 1.10 (t, 6H, CH_3)
7d	Light yellow powder	>220	ν 2974.72 (N-H), 1656.13 (C=N), 1215.77 (C-N)	7.97 (s, 1H, N-H), 8.21 (d, 2H, Ar-H), 7.75 -7.78 (m, 4H, Ar-H), 7.68 (d, 2H, Ar-H), 7.58 - 7.60 (m, 4H, Ar-H), 5.57 (s, 1H, Ar-H), 3.55 (q, 4H, CH_2), 1.17 (t, 6H, CH_3)

As observed, the synthesized benzimidazoles have relatively modest yields most likely due excess aryl aldehyde used as starting material for synthesis and the unseemly solvent choice for recrystallization [21,22], which might influence the optimal crystallization of the products. Albeit limited product yields were noted, it still demonstrated the advantages of solvent-free process over the conventional ones such as no reaction media to collect, purify, and recycle. Products are rapidly formed in minutes and are often pure enough to avoid using column chromatography. The process can made using simple set up with minimal energy requirement. Moreover, this study was focused only on the synthesis and subsequent bioactivity screening hence optimization of reaction parameters was no longer performed.

3.3 Brine shrimp lethality assay (BSLA) screening of synthesized 2-aryl-1H-benzimidazoles

The recrystallized products were subjected to BSLA testing. Each compound was tested at 10, 100, and 1000ppm concentrations. The number of dead and alive brine shrimps were counted after 6 and 24 hours. Due to the highly nonlinear mortality data obtained, the Miller and Tainter method [16–19], a modified Probit analysis was utilized to calculate the LC_{50} for 6 hours and 24 hours corresponding to the acute and chronic toxicity values (Table 3).

Table 3 BSLA results of synthesized 2-Aryl-*1H*-benzimidazoles.

Product	Acute toxicity LC_{50} (6h) (ppm)	Chronic toxicity LC_{50} (24h) (ppm)
5a	>1000.00	>1000.00
5b	43.42	<10.00
5c	>1000.00	283.39
5d	>1000.00	73.93
6a	955.86	<10.00
6b	>1000.00	<10.00
6c	>1000.00	11.35
6d	405.90	<10.00
7a	>1000.00	195.02
7b	>1000.00	<10.00
7c	>1000.00	145.84
7d	>1000.00	<10.00

3.4 Qualitative Structure-BSLA Activity Correlation

As a guide for the qualitative structure activity correlation, the benzimidazole scaffold below (Figure 5) will serve as basis of comparison among the toxicity values determined for the synthesized benzimidazoles.

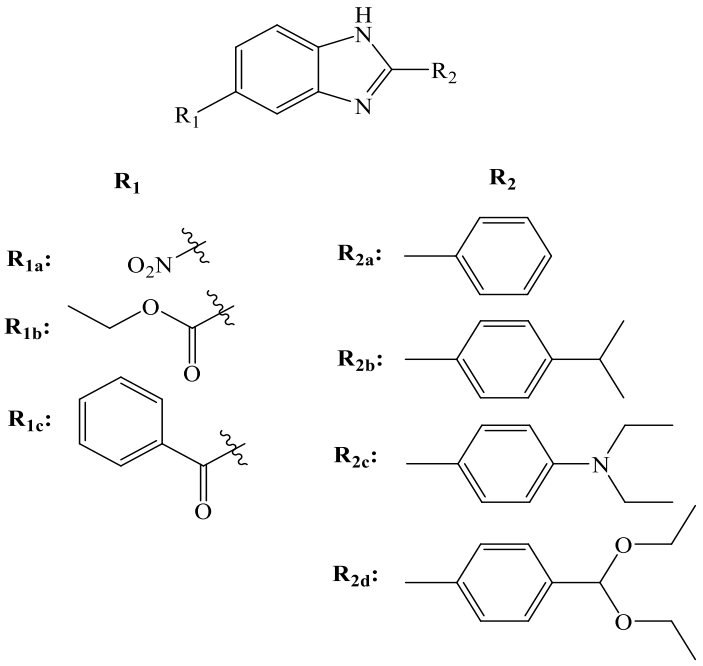


Figure 5 Molecular scaffolds of the synthesized benzimidazoles.

Ranked bioactivity values of the benzimidazoles are presented in Table 4. Structural effects of the substituents (R_1 and R_2) on the toxicity values of the molecular scaffolds were evaluated on a qualitative basis only.

Table 4 Qualitative correlation of structural effects and activity of 2-Aryl-1*H*-benzimidazoles.

Rank	Product	Toxicity values		Benzimidazole Substituents	
		LC_{50} (24h) (ppm)	LC_{50} (6h) (ppm)	R_1	R_2
1	5b	<10.00	43.42	R_{1a}	R_{2b}
2	6d	<10.00	405.90	R_{1b}	R_{2d}
3	6a	<10.00	955.86	R_{1b}	R_{2a}
4	6b	<10.00	>1000.00	R_{1b}	R_{2b}
	7b	<10.00	>1000.00	R_{1c}	R_{2b}
	7d	<10.00	>1000.00	R_{1c}	R_{2d}
5	6c	11.35	>1000.00	R_{1b}	R_{2d}
6	5d	73.93	>1000.00	R_{1a}	R_{2c}
7	7c	145.84	>1000.00	R_{1c}	R_{2c}
8	7a	195.02	>1000.00	R_{1c}	R_{2a}
9	5c	283.39	>1000.00	R_{1a}	R_{2c}
10	5a	>1000.00	>1000.00	R_{1a}	R_{2a}

The effect of the R_1 substituents on the synthesized monosubstituted benzimidazoles on the observed toxicity is shown in Figure 6. Apparently, the enhancing effects of the R_1 substituents on the toxicity of the 2-Aryl-1*H*-benzimidazoles is opposite to the trend in increasing electron withdrawing effects of R_{1a}, R_{1b}, and R_{1c}.

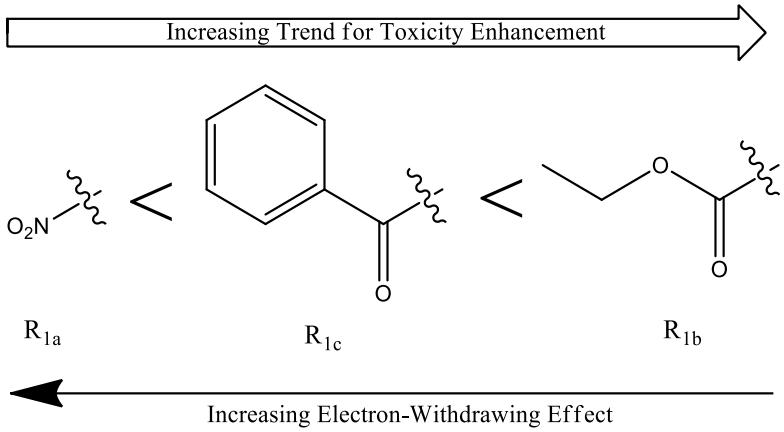


Figure 6 Structural effects of R_1 substituents on toxicity.

Doing a parallel analysis on the R_2 substituents, the increasing order for enhancing toxicity is demonstrated in Figure 7. The enhancing capacity of the R_2 substituents on the toxicity of the 2-Aryl-1*H*-benzimidazoles is similar with the R_1 substituents having an opposite effect as increasing electron donating effects of R_{2b}, R_{2c}, and R_{2d} apart from R_{2a} having the slightest electron donating effect among the R_2 substituents as shown in Table 3.

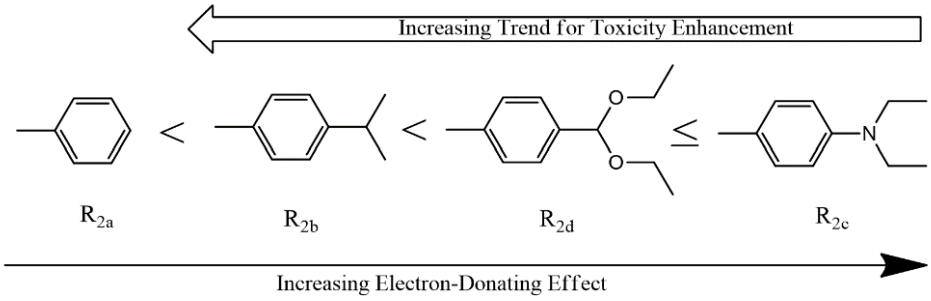


Figure 7 Structural effects of R_2 substituents on toxicity.

Combining the effects of R_1 and R_2 substituents, it was observed that if both electron-donating ability of R_2 and the electron-withdrawing effect of R_1 were placed to a minimum, the toxicity of 2-Aryl-1*H*-benzimidazole is enhanced. This correlation may be due to the more well-balanced electron density on the imidazole core which is

known in literature as the bioactive function of benzimidazoles. This increased stability electron density on the imidazole scaffold (Figure 8) may boost specific interactions with receptor molecules responsible for the toxic effects against the test organism *Artemia salina* in the BSLA. Moreover, the detected low-level toxicity on benzimidazoles product with R_{2a} substituent may be attributed to the disturbance of electron density of imidazole core especially if very strong electron withdrawing substituents were present in R_1 position.

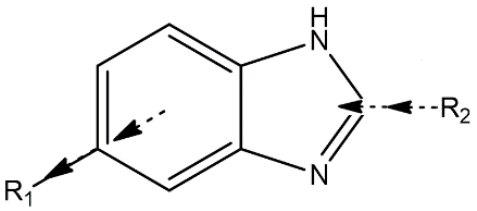


Figure 8 Repressed electron-donating capacity of R_2 corroborated with suppressed electron-withdrawing effect of R_1 leading to enhanced toxicity of 2-Aryl-1*H*-benzimidazole.

4. Conclusions

Twelve 2-Aryl-1*H*-benzimidazole derivatives were successfully synthesized *via* a solvent-free room-temperature approach employing a P_2O_5/SiO_2 catalytic system. Modest yields were obtained for the target compounds, but this may be increased by preforming further optimization of reaction conditions. Ensuing toxicity testing *via* BSLA of the benzimidazoles revealed the excellent potency of the synthesized products in terms of bioactivity. Specifically, eight (5b, 5d, 6a, 6b, 6c, 6d, 7b, and 7d) out of the twelve benzimidazoles exhibited chronic toxicity (LC_{50} 24h) values below 100 ppm. Qualitative structure-activity relation revealed that the toxicity of the prepared benzimidazoles is well correlated with the electron-donating/electron-withdrawing effects of the substituents present. Overall, increased stability on the electron density on the imidazole function effected by electron donation enhances the toxicity of the described benzimidazoles.

5. Acknowledgements

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