FUNGICIDAL ACTIVITY OF SOME INDOLE DERIVATIVES

Ahmed S. Abdel-Aty

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ABSTRACT

Fungicidal activity of six prepared indole derivatives in addition to indole-3-acetic acid and indole-3-butyric acid was in vitro evaluated against Fusarium calmorum, Pythium debarianum, Rhizoctonia solani and Macrofomina phaseoli fungi. The toxicity appeared to be a function of both the treated fungus and the tested concentration. IC_{50} values were calculated and the structure activity relationship (SAR) was illustrated. 2-phenylindole and 1-acetylindole-3-butyric acid exhibited persuasive fungicidal activities, so their effects were examined in vivo against polyphenoloxidase (PPO), peroxidase (PO), DNA and RNA contents and the fungal sugar contents. Polyphenoloxidase in R. solani systematically responded to 2-phenylindole concentrations with IC_{50} value of 80.3 µg/ml. 1-acetylindole-3-butyric acid was more effective than 2-phenylindole with IC_{50} values of 41.5 and 80.2 µg/ml comparing with 87.6 and 117.1 µg/ml on F. calmorum and M. phaseoli, respectively. While against P. debarianum, the enzyme was inhibited by 1-acetylindole-3-butyric acid with IC_{50} equaled 45.6 µg/ml. Effect of the tested compounds on peroxidase was differed among the tested fungi. The peroxidase extracted from P. deparianum was less inhibited. Changing in sugar, DNA and RNA contents of the tested fungi were exhibited that explains disturbance in the fungal cell physiology and developing deformed and dead cells.

INTRODUCTION

Different researches are directed to evaluate new compounds as effective fungicides to face continuous fungal infections. Indole nucleus occupies a position of major importance as antimicrobial agents in the vast heterocyclic structural space. Combination of indole-3-acetic acid (IAA) as the most active endogenous auxin involving in various physiological processes in higher plants at 100 µg/ml with *Cryptococcus laurentii* suppressed blue and gray mold infections (*Penicillium expansum* and *Botrytis cinerea*) on pear fruit more than *C. laurentii* alone (Yu and Zheng, 2007). It affected the dry rot causative pathogen, *Gibberella pulicaris* suppressing infection of wounded potatoes, optimally when combined with phenylacetic acid and tyrosol (Slininger *et al.*, 2004). It reduced spore germination, mycelial dry weight and protein content of tomato wilt pathogen *Fusarium oxysporum lycopersici* matching with its concentration elevation preventing any chance for disease incidence by soil pathogens when applied *in vivo* to the soil (Sharaf and Farrag, 2004). It also inhibited the mycelial growth of the *Macrophomina phaseolina in vitro* and appeared quite effective to reduce the charcoal rot disease both in field and greenhouse (Kumar *et al.*, 2007). Its 5-methoxy derivative showed antifungal activity against *F. oxysporum*, *Rhizoctonia solani* and *Coprinus comatus* and it was the most potent among pineal products against *Agrobacterium tumefaciens* (Wang and Ng, 2002). Several indole derivatives showed antimicrobial activity as 1H-indole-4,7-diones exhibited potent antifungal activity against *Candida krusei*, *Cryptococcus neoformans*, and *Aspergillus niger* (Ryu *et al.* 2007). The methanol extract from calli seeds using Gamborg's B5 basal media supplemented with indole-3-butyric acid (1.0 ppm), 6-benzylaminopurine (N(6)-benzyladenine) (1.0 ppm), and sucrose (2.5%) had great potential antimicrobial activities against 23 bacteria and 15 fungi and yeast species tested (Gulluce *et al.*, 2003). 5-nitro-2-phenyl-1H-indole as well as 2-aryl indole derivatives with ortho substitution on the phenyl ring inhibited the N or A efflux pump in the human pathogenic bacteria, *Staphyllococcus aureas* (Samosorn *et al.*, 2005).

So in the present study, six indole derivatives were prepared, purified and identified by NMR and Mass spectroscopy measurements. These compounds, in addition to indole-3-acetic acid and indole-3-butyric acid were studied for their fungicidal effect against the damping off fungi *Fusarium calmorum, Rhizoctonia solani, Pythium debarianum* and *Macrofomina phasoli* that causes post harvest fruits rotting. *In vivo* inhibition rate by the active derivatives were measured on polyphenoloxidase, peroxidase, DNA, RNA and sugar contents in the treated fungi.

MATERIALS AND METHODS

I- Preparation of the tested compounds:

Both indol-3-acetic acid GRG, El-Gomhouria Drug Company, batch No 971381, Code No L 17070 and indol-3-butyric acid, Sisco Research Laboratories PVT. LTD, Mumbai, India, Batch No T828452 were purchased from El-Gomhouria Drug Company, Egypt. Other tested derivatives were prepared and identified (Table 1, Figure 1) as follow:

1- Benzoyl indole-3-acetic acid

Indol-3-acetic acid (1 gm, 0.0057 mole) was dissolved in 40 ml of 10% aqueous sodium hydroxide with stirring for 15 minutes at room temperature. Benzoyl chloride (2 ml) was added in portions within 20 minutes (Benson *et al.*, 1952). The content was acidified with concentrated hydrochloric acid to give

Department of Pesticide Chemistry, Faculty of Agriculture, 21545-El-Shatby, Alexandria University; Alexandria, Egypt

a white precipitate that was filtered. The inorganic salts were removed by 2.0 liters of water. The crude yield was recrystallized from 95% aqueous ethanol to give pure 1-benzoyl indole-3-acetic acid (0.75 gm, 47.0%) with 117-118.5° C melting point. ^{1}H NMR (DMSO-d6) showed the broad carboxylic proton at δ 12.85. Benzoyl protons appeared at δ 7.94, 7.97 and 7.98 for *meta*, *para* and *ortho* protons whenever, signals distributed at δ 7.45 (1H, s, 2-H), δ 7.47 and 7.58 (2H, d, 5-H and 6-H, J = 5.1 and 5.1 Hz) and δ 7.62 (2H, d, 4-H and 7-H, J = 5.1 Hz) are due to indole protons. These multiplicities of signals are referred to C4-C5, C4-C6, C5-C6 and C5-C7 coupling. Signal at δ 3.85 (2H, s, CH_2) is referred to the aliphatic CH_2 group. EI-Mass spectrum, loss of the carboxylic group from the parent molecular ion gives 234 m/z fragment, which cleaved at C=O producing to $C_6H_5^+$, $C_6H_5CO^+$ ions and the base peak at 77, 105 and 130 m/z, respectively. When the parent compound deprived of the carboxylic group firstly gives fragment at 175 m/z that bereft of benzoyl ion to give the base fragment. Fragments at 122, 207 and 117 m/z are produced through ring fission and loss of the carboxylic group.

1-Acetyl indole-3-butyric acid

Indole-3-butyric acid (1.0 gm) was refluxed with 50 ml of acetic anhydride and 2 ml of glacial acetic acid for 40 minutes and poured into ice with stirring. The mixture was kept overnight at 6° C. Yellowish white precipitate was filtered and washed well with water. Recrystallization of the yield from water: ethanol (1:3) gave 0.903 gm (74.8%) of 1-acetyl indole-3-butyric acid, m.p. 165-166° C. ^{1}H NMR spectrum (DMSO-d6) exhibited the carboxylic proton at δ 11.55 whereas, the aliphatic methyl group singlet at 2.49 δ. Indole ring protons arranged at δ 7.05 (1H, d, 2-H, J = 1.0 Hz), δ 7.32 (2H, dd, 5-H and 6-H, J = 8.1 and 8.1 Hz) and δ 7.65 (2H, d, 4-H and 7-H, J = 9.0 Hz). The aliphatic CH_2 groups appeared their signals at δ 2.94 (2H, t, 1-2H), δ 2.50 (2H, m, 2-2H) and δ 2.15 (2H, t, 3-2H). This multiplicity is due to coupling among propyl protons. Mass spectrum revealed fragmentation of the molecular ion at m/z 245 by loss both the acetyl and carboxyl groups to 157 m/z fragment, which loses one or two CH_2 groups to 143 m/z and 129 m/z (base peak), respectively. Lack of either CH_3 and COOH groups lead to 185 m/z fragment that loses either C_2H_4 group to 157 m/z fragment or CNO group (through the ring fission) to 143 m/z fragment.

1-Benzoyl indole-3-butyric acid

Indol-3-butyric acid (1 gm, 0.005 mole) was mixed with 40 ml of 10% aqueous sodium hydroxide and 30 ml of 95 % aqueous ethanol. The solution was stirred for 5 minutes at room temperature, it was turned yellow. Benzoyl chloride (3 ml) was added in portions through vigorous stirring. The reaction mixture became colorless. The stirring was continued for 15 minutes. Few crystals were appeared and filtered off. The filtrate was acidified with diluted hydrochloric acid and incubated below 0°C overnight. An oily material was produced. The product was recovered in 50 ml of 95 % aqueous ethanol and mixed well. Concentration of the final solution to 20 ml under reduced pressure following with keeping at 6° C overnight lead to white powder. Recrystallization of this product from aqueous ethanol gave 1.14 gm (75.4%) of 1-benzoyl indole-3-butyric acid with 113-114° C melting point. ^{1}H NMR spectrum (DMSO-d6) showed the carboxylic proton at δ 12.90. Signals of the benzoyl protons overlapped at δ 7.95, 7.97 and 7.99 for meta, para and ortho protons. Protons of the distributed at δ 7.45 (1H, s, 2-H), δ 7.48 and δ 7.55 (2H, d, 5-H and 6-H, J = 5.1 and 5.1 Hz) and δ 7.60 (2H, d, 4-H and 7-H, J = 5.1 Hz). The aliphatic chain arranged its signals at δ 4.45 (2H, t, 1-2H), δ 4.05 (2H, m, 2-2H) and δ 2.3 (2H, t, 3-2H). These chemical shifts were downfield shifted in comparison to the acetyl indole-butyric acid due to phenyl ring presence instead of methyl group. Mass spectrum exhibited that loss of the carboxylic proton to give M-1$^+$ at 306 m/z. Release the benzoyl group and subsequently the phenyl ion as m/z 105 and 77 fragments is the characterizing step. The remained moiety was protonated to 203 m/z fragment, which loses COOH group following by loss C_2H_4 group to m/z 158 and 130 fragments, respectively. Ring cleavage leads to benzamide group protonated to m/z 122 fragment as the base peak.

2-Phenylindole

Phosphorus pentoxide (P_2O_5) (160 gm) was completely dissolved in 100 ml of ortho-phosphoric acid with heating as an alternative reagent in spite of the commercial poly phosphoric acid (Vogel, 1976). Phenyl hydrazone was firstly prepared as a starting material by mixing acetophenone (20 gm, 0.167 mole) with 18 gm (0.167 mole) of phenyl hydrazine and 60 ml of ethanol. The reaction mixture was acidified with 1.0 ml of glacial acetic acid and warmed for 10 minutes. After half cooling, 40 ml of 10 % sodium acetate solution was added and the mixture was completely cooled. The produced yellowish phenyl hydrazone was filtered and washed with 500 ml of diluted hydrochloric acid and 100 ml of ethyl alcohol. The product turned to white solid with melting point of 105-106°C. The produced phenyl hydrazone (22 gm) was introduced to 100 ml of the prepared poly-phosphoric acid in a 400 ml beaker and the mixture was heated to 150 °C on a hot plate with continuous stirring (the reaction with effervescence started at 55 °C). After cooling to 110 °C, cold water was added and a creamy precipitate was filtered under vacuum, refluxed with 300 ml of ethanol and filtered under vacuum in a pre-heated buchner funnel. The residue

was washed with ethanol and the combined filtrates were concentrated to give 16 gm (71%) of crude yield with 187-190°C melting point. Recrystallization from 95% aqueous ethanol (10 ml/gm) in presence of little charcoal gave the pure 2-pheny indole with 189-190°C melting point, 93% recovery (Ref. 188-189°C). ^{1}H NMR spectrum (DMSO-d_6) explained the structure as the singlet signal at 11.55 δ is due to N-H proton. The C-H proton of indole ring appeared at δ 6.9 (1H, s, 3-H), whenever the two protons at C4 and C7 appeared at 7.89 δ (2H, d, J = 8.67 Hz due to C4-C5 and C6-C7 coupling). Two doublet signals centered at δ 7.4 and δ 7.5 (2H, dd, J = 8.67 and 8.67 Hz due to coupling with C4 and C7) are due to protons at C5 and C6 atoms. All signals are downfield shifted with 0.5- 1.0 δ than that of the un-substituted indole ring due to presence of the phenyl ring. Signals of the substituted phenyl ring protons appeared at the region 7.0 to 7.34 δ, at δ 7.03 (2H, dd, *ortho*-2H, J = 8.3 Hz due to coupling between ortho and meta protons, δ 7.14 (1H, d, *para*-1H, J = 8.3 Hz) due to coupling between para and meta protons and 7.31 δ (2H, d, *meta*-2H, J = 8.3 Hz) due to coupling between protons at (ortho or para) and meta positions. Mass spectrum showed the molecular ion (M^+) and its protonated and de-protonated fragments ($M+1^+$ and $M-1^+$) at 193, 194 and 192 m/z. The molecular ion peak appeared as the base peak. Loss of C_2H_4 leads to fragment at 165 m/z. Fragments at m/z 89 and 63 are produced by loss a benzonitril ion ($C_6H_5CN^+$) from m/z 192 and 165 fragments through the ring fission, respectively. Fragment at m/z 90 and 89 are produced through the same step.

1-Acetyl-2-phenylindole

The prepared 2-phenylindole (1.34 gm, 0.007 mole) was acetylated by refluxing with 60 ml of acetic anhydride and 1.0 ml of acetic acid for 40 minutes, followed by pouring into ice water and filtration of the produced precipitate under reduced pressure. The yield was washed several times with water and recrystallized from 95% aqueous ethanol to give pure 1-acetyl-2-phenylindole (0.45 gm, 46.1%) as an off white powder with melting point equals 179-180.5°C. ^{1}H NMR spectrum (DMSO-d_6) showed the methyl protons signal at 3.37 δ. Aromatic protons signals were distributed as in case of 2-phenylindole as the C-H proton of indole ring appeared at δ 6.8 (1H, s, 3-H), δ 7.47 and δ 7.54 (2H, dd,5-H and 6-H, J = 8.67 and 8.67 Hz), δ 7.87 (2H, d, 4-H and 7-H, J = 8.67 Hz) The phenyl signals appeared at δ 7.02 (2H, dd, *ortho*-2H, J = 8.3 Hz), 7.11 (1H, d, p-1H, J = 8.3 Hz), δ 7.3 (2H, d, m-2H, J = 8.3 Hz), δ 3.37 (3H, s, COCH3). Mass spectrum showed the molecular ion (M^+) and its protonated fragments ($M+1^+$) at 235 and 236 m/z. The molecular ion gives the 192 m/z fragment ($M-CH_3CO)^+$ and its protonated ion at 193, whenever loss of only the aliphatic methyl group gives 220 m/z fragment ($M-CH_3)^+$ that produced 192 m/z fragment through the ring fission. Ring fission of the resulted 2-phenylindole fragment produced the fragments at m/z165, 103 and 91 by ring fission path way by loss of C_2H_4 and benzonitril molecules.

1-Benzoyl-2-phenylindole

The prepared 2-phenylindole (3 gm, 0.0155 mole) was dissolved in 70 ml of 5% aqueous sodium hydroxide. Benzoyl chloride (5 ml, 0.049 mole) was added at room temperature with vigorous stirring in 20 minutes. The solution became warm and changed to violet in color. After keeping in freezer for 48 hours, crystalline powder was formed and separated under vacuum. The yield was washed with water to remove the alkaline and recrystallized from ethanol in presence of charcoal to give the pure 1-benzoyl-2-phenylindole (3.85 gm, 83.4%) with 106-107°C melting point. ^{1}H NMR spectrum (DMSO-d_6), the C-H proton of indole ring appeared at δ 6.9 (1H, s, 3-H), whenever the two protons at C4 and C7 appeared at δ 7.97 (2H, d, J = 5.33 and 1.67 Hz due to C4-C5 and C6-C7 coupling). Two doublet signals centered at δ 7.88 (2H, dd, J = 8.0 and 1.33 Hz due to coupling with C4 and C7) are due to protons at C5 and C6 atoms. All of the explained signals are downfield shifted than that of un-substituted indole ring due to presence of the phenyl and benzoyl moieties. Signals of the substituted phenyl ring protons appeared at the region 7.0 to 7.33 δ. At δ 7.02 (2H, dd, *ortho*-2H, J = 7.6, 1.0 Hz due to ortho-meta and ortho-para coupling), δ 7.1 (1H, d, *para*-1H, J = 8.0 Hz due to coupling between protons at para and meta positions) and δ 7.31 (2H, d, *meta*-2H, J = 7.66 Hz due to coupling between protons at ortho or para with meta positions). Signals at δ 7.43 (2H, dd, meta-2H, J = 5.33 and 3.66 Hz), δ 7.48 (1H, dd, para-H, J = 8.3 and 2.7 Hz) and 7.43 δ (2H, dd, ortho-2H, J = 8.3 Hz) are referred to meta, para and ortho protons of benzoyl group, respectively. Mass spectrum showed the main fragmentation pathway is cleavage of carbonyl group bonds to phenyl and the benzoyl ions at 77 and 105 m/z, besides the remained fragment protonated to the 2-phenylindole ion at 193 m/z, the base peak fragment. The compound through fission of the indole ring gives fragment at 122 m/z. Fragment at 193 m/z was fragmented in the same manner of 2-phenylindole.

II- Tested fungi:

Four species of plant pathogenic fungi: *Fusarium calmorum*; *Pythium debarianum*, *Rhizoctonia solani* and. *Macrofomina phaseoli* were chosen because as economical fungi threating several crops. These species were provided from Plant Pathology Department, Faculty of Agriculture, Alexandria University, El-Shatby, Alexandria, Egypt.

III- Methods of Testing:

A. Fungitoxic effects:

Measurements were carried out by using radial growth test according to the conventional method reviewed by Torgeson (1967) as follow: A definite volume of the well-known Czapek-Dox medium (12 ml) containing agar (4.5 gm/100 ml water) was sterilized in conical flasks. Citrate-Phosphate buffer solution (3 ml) was autoclaved separately; both solutions were mixed in a sterile conical flask. The tested compounds dissolved in dimethyl-sulfoxide (DMSO) were finally added to be 1, 10, 50, 100, 200 and 500 µg/ml. The contents of each flask (36 ml) were distributed in three sterilized petri-dishes and considered as one treatment. All additions were done under aseptic conditions. After solidification, the inoculum disc (7 mm in diameter) of each tested fungus was located in the center of the petri-dish. Control in the presence of the calculated volume of dimethylsulfoxide only to be 1 % as its final concentration was concurrently conducted. The results were recorded by measuring the diameter of the hyphal growth when the growth of the untreated fungi completely covered the surface of petri-dish. The inhibition percent of the hyphal growth were calculated according to the following formula (Topps and Wain, 1957).

$$\% \text{ Inhibition} = 100\ [(C-T) / C]$$

C = Average radial hyphal growth in control
T = Average radial hyphal growth in treatment

IC_{50} values as µg/ml (the concentration caused 50% inhibition in the hyphal growth) were calculated for each compound and fungus using probit analysis method (Finney, 1971). Data were compared with that of the technical grade of metalaxyl 98% (Radomil), methyl-N-(2,6-dimethylphenyl-N-methoxyacetyl)-DL-alaninate as standard fungicide

B. Biochemical effects:

Biochemical interaction measurements were conducted *in vivo* by the determination of polyphenoloxidase and peroxidase activities on the tested fungi using liquid medium experiments as follows: The well known Czapek-Dox medium were prepared in conical flasks containing the tested concentrations to be 0, 0.1, 0.25, 0.5, 1 and 2 of IC_{50} values of 2-phenylindole and 1-acetylindole-3-butyric acid on each fungus. An inoculum disc of the tested fungus was located on the surface of the mediun in each flask. When the hyphal growth in untreated flask was completely grown, the contents of each flask were filtered under vacuum and used for determining the enzymes activities.

1-Polyphenoloxidase activity (PPO):

The samples of the hyphal growth were homogenated with borate buffer (pH 9.0) and centrifuged at 4000 rpm for 15 minutes. The supernatant was used for determining polyphenoloxidase activity (PPO) at 575 nm (Broesch, 1954).

2- Peroxidase activity (PO):

The samples of the hyphal growth were homogenated with phosphate buffer (pH 6.0) and centrifuged at 4000 rpm for 15 minutes. The supernatant was used to measure peroxidase activity (PO) 470 nm (Fehrmaun and Dimond, 1967).

The homogenates were measured for their mg protein contents (Lowry *et al.*, 1951) and the specific activities of all treatments were determined. Inhibition percents were calculated according to (Topps and Wain, 1957).

3- DNA and RNA content:

The dried crushed hyphal growth (0.1 gm) was extracted with 5 ml of perchloric acid (0.5M) on a boiling water bath for 20 minutes. After cooling both DNA and RNA contents were determined in mg/liter using spectrophotometer at 270 nm and 290 nm, respectively, according to Stoev and Makarova (1989). All previous spectroscopic measurements were done using Nicolet 100 UV-VIS Spectrophotometer, Thermo Electron Corporation.

4- Effect on sugar contents:

The air-dried hyphae of *M. phaseoli* or *R. solani* (0.5 gm) was blended with 10 ml of 80% aqueous ethanol and 1 ml aliquot of the resulted extract was used for determination of total soluble sugars (T.S.S), reducing sugars (R.S) and non-reducing sugars (non-R.S) using the picric acid method (Thomas and Dutcher, 1924). The absorbance of the resulted color was measured at 540 nm on Unico-1200 Spectrophotometer against a standard solution of glucose. The sugar contents were determined as mg/gm air dried fungal hyphae.

IV- Statistical analysis:

Inhibition percents in hyphal growth were analyzed using the analysis of variance (ANOVA) and Student-Newman-Kules Test. IC_{50} values were calculated by using probit analysis method (Finney, 1971)

RESULTS AND DISCUSSION

I- *In vitro* fungitoxic effects:

Fungitoxic effects of the tested indole derivatives and metalaxyl (radomil) as the used standard fungicide were recorded as IC_{50} values in Table (2). Against *F. calmorum*, derivatives of 2-phenylindole were more effective than the standard

fungicide with IC_{50} values ranged from 67.4 to 99.9 µg/ml. 1-Acetylindole-3-butyric acid appeared to be the most active with IC_{50} value equaled 26.6 µg/ml. The other prepared derivatives were less effective than the standard fungicide (metalaxyl). *M. phaseoli* was affected in the same array with less toxicity degree. 1-Benzoyl-2-phenylindole slackened in its effect to be less than the standard fungicide. Fungicidal activity was increased against *P. debarianum* in all cases in comparison to *F. calmorum*. The derivatives: 2-phenylindole, 1-acetyl-2-phenylindole and 1-benzoyl-2-phenylindole inhibited its hyphal growth with IC_{50} values equaled 17.7, 15 and 81 µg/ml, respectively in comparison to 211 µg/ml of the standard fungicide. 1-Acetylindole-3-butyric acid was very active with IC_{50} value equaled 19 µg/ml. *R. solani* appeared to be more tolerant than the other tested fungi for all of the tested compounds including the standard fungicide. Both 2-phenylindole and its 1-acetyl- derivative showed high effects with IC_{50} values equaled 34.6 and 37.5 µg/ml. 1-Acetylindole-3-butyric acid exhibited less fungitoxic activity than on the other fungi but it is still more active than metalaxyl, followed by 1-benzoyl-2-phenylindole with 117 and 122.2 µg/ml, respectively. From the mentioned results, fungicidal activity proved to be a function of both the treated fungus and the derivative structure. Regarding the tested fungi, *P. debarianum* was the most sensitive, followed by *F. calmorum*, *R. solani* and *M. phaseoli*. Their hyphal growth was inhibited with Mean ± SE equaled 35.52 ± 2.16, 30.02 ± 1.99, 28.02 ± 1.66 and 25.31 ± 1.49 µg/ml, respectively with significant differences. Regarding the structure activity relationship, it was noticed that acylation of the natural auxin enhanced its fungicidal activity as substitution with a 1-benzoyl moiety in indole-3-acetic acid (IAA) slightly increased the activity although in case of indole-3-butyric acid (IBA) it had no significant effect. Acetylation of IBA strongly multiplied the activity against all tested fungi. Replacing the 3-aliphatic chain with 2-phenyl moiety firmly improved the toxicity against all the tested fungi. While benzoylation of 2-phenylindole decreased its activity, acetylation maintained its toxicity high. Based on statistical analysis, 1-acetylindole-3-butyric acid, 2-phenylindole, 1-acetyl-2-phenylindole and 1-benzoyl-2-phenylindole exhibited their inhibition with Mean ± SE equaled 44.65 ± 3.91, 43.07 ± 3.32, 42.36 ± 3.38 and 31.02 ± 2.76 µg/ml, respectively surpassing the standard fungicide with 29.05 ± 2.46 µg/ml. The other structures were less effective than the standard fungicide.

II- Biochemical Effects:

1- Effects on polyphenoloxidase and peroxidase

Results of both 2-phenylindole and 1-acetylindole-3-butyric acid effects on polyphenoloxidase and peroxidase activities are shown in Tables (3 and 4). Polyphenoloxidase activity in *R. solani* systematically responded to 2-phenylindole with IC_{50} equaled 80.3 µg/ml. 1-Acetylindole-3-butyric acid inhibited the enzyme activity with 39% at the lowest concentration. This effect was decreased to 0% at 0.5 IC_{50}, followed by activation with 78.9 and 84% of control at 1.0 and 2.0 IC_{50} values, respectively. In all other cases, 1-acetylindole-3-butyric acid was more effective than 2-phenylindole in inhibition of polyphenyloxidase with 41.5 and 80.2 µg/ml IC_{50} values comparing with 87.6 and 117.2 µg/ml, respectively in case of *F. calmorum* and *M. phaseoli*. While *P. debarianum* enzyme activity was inhibited by 1-acetylindole-3-butyric acid with IC_{50} equaled 45.6 µg/ml, 2-phenylindole enhanced it with activating concentration of 50% (AC_{50}) equaled 35.1 µg/ml. Regarding the effect on peroxidase, in *R. solani* it was activated with AC_{50} equaled 14.5 and <11.7 µg/ml in case of 2-phenylindole and 1-acetylindole-3-butyric acid, respectively. Both the two compounds exhibited narrow ranged inhibitory effects against the enzyme from *P. debarianum*. While in *M. phaseoli*, the enzyme was activated systematically with 1-acetyl indole-3-butyric acid with AC_{50} < 5.9 µg/ml, 2-phenylindole affected it from -39 to 54.3 % inhibition regularly with increasing its concentration. It affected the activity of *F. calmorum* enzyme from 85.0 to -115.3 % inhibition within its concentration range. This activity was inhibited with 2-phenylindole with IC_{50} equaled 49.9 µg/ml. (Table 4). From the previous data it was noticed that the effect was a function of both the tested fungus and the used compound. In all cases the enzyme activity was systematically affected with the tested concentration. Peroxidase from *P. deparianum* was less sensitive as it was affected in narrow range of inhibition or activation, while it was different in case of polyphenoloxidase. The effect also changed regarding the tested compound which maybe due to the chemical structure differences like the type and position of the substituent on the indole ring.

2- Effect on DNA and RNA contents:

Effect of 2-phenylindole on RNA and DNA contents in each tested fungus at several rates of their IC_{50} values are recorded in Table (5). Both RNA and DNA molecules are related to each other and RNA molecules are formed in the nucleus by transcription of genetic information encoded in the sequence of DNA basis; so the results obtained were exhibited in systematic response with both RNA and DNA content. In *F. calmorum*, RNA and DNA contents as compared with control (51.5 and 49.5 mg/liter) were found to be reduced at all the tested IC_{50} rates of 2-phenylindole. This reduction was increased with increasing the tested concentration to 0.5 IC_{50} then RNA content was dramatically increased to 26.3 and 24.7 mg/liter and DNA content was increased to 25.3 and 23.8 mg/liter at 1 and 2 IC_{50}. On the other hand, RNA and DNA

contents in *R. solani* highly increased more than control and reached to the maximum peak of increase at 1.0 IC_{50} of 2-phenylindole. RNA and DNA contents in *M. phasoli* were reduced to less than 50% of control at all the tested rates. They changed from 8.3 to 5.9 and from 8.0 to 5.7 mg/liter comparing with 16.1 and 15.5 mg/liter of control. These contents of *P. deparianum* behaved the same trend of these in *M. phasoli* changing from 30.6 to 11.4 and from 29.4 to 10.9 mg/liter at the tested concentrations comparing with 32.2 and 31 mg/liter of control in nonsystematic arrangement.

1-Acetylindole-3-butyric acid affected both RNA and DNA contents differently according to the tested fungi and concentration. It reduced them in *F. calmorum* in systematic arrangement at all the tested IC_{50} rates comparing with control. While RNA and DNA contents in *M. phaseoli* were decreased by increasing the tested rate with systematical arrangement. This reducing effect was noticed in all fungi. The RNA and DNA contents were reduced from 45.4 to 20.3 and 43.6 to 19.5 mg/L in case of *F. calmorum*, they were reduced from 12.9 to 6.7 and from 11.7 to 6.5 in case of *M. phaseoli* comparing with 51.5, 49.5, 16.1 and 15.5 of their control, respectively. While the contents from *P. debarianum* were decreased untill 0.5 IC_{50} and increased again at the two highest concentration rates, they were systematically increased with increasing the concentration in case of *R. solani* Table (6). General descriptive analysis proved that 2-phenylindole affected *M. phasoli* significantly greater than *P. debarianum* with (8.42 ± 0.86 and 25.05 ± 1.84) and (8.08 ± 0.83 and 24.1 ±1.78) mg/liter means ± SE of RNA and DNA contents. Although there was no significant difference between *R. solani* and *F. calmorum*, they differed significantly from the other tested fungi with (29.2±2.55 and 29.23±0.55) and (28.12±2.45 and 28.16±0.42) mg/liter of RNA and DNA contents. The same arrangement was exhibited in treatment with 1-acetylindole-3-butyric acid except achieving a significant difference among all the tested fungi. RNA contents were 10.57 ± 0.78, 23.01 ± 1.61, 28.57 ± 1.07 and 34.31 ± 2.61 mg/liter, while DNA contents were 10.09 ± 0.73, 22.07 ± 1.53, 27.52 ± 1.01 and 33.0 ± 2.51 mg/liter in case of *M. phasoli*, *P. debarianum*, *R. solani* and *F. calmorum*, respectively. However, the changes in DNA and RNA contents in the tested fungi that may develop deformed and dead cells.

3- Effect on sugars contents:

Effect of the tested compounds on sugar contents are presented in Table (7). Comparing with the untreated fungus, all the sugar types of *M. phasoli* were decreased at all 2-phenylindole concentrations with non-systematic arrangement. At 0.1 IC_{50} of 1-acetylindole-3-butyric acid, its sugar contents were drastically reduced, followed by non systematic increasing with increasing the treated concentration in case of non-reduced sugars , while the two other sugars were systematically increased. *R. solani* sugars contents were strongly multiplied at 0.1 and 0.25 IC_{50} concentrations. At 0.5 IC_{50}, a firm decrease was exhibited in both reduced and non-reduced sugars. This reduction was increased at 1.0 IC_{50}. This effect was differed from the effect of 1-acetylindole-3-butyric acid as both reduced and non-reduced sugars were systematically decreased with increasing the tested concentration. Both the reduced and non-reduced sugars as well as the total soluble sugars were *in vivo* affected with the two studied compounds in a treated fungus and concentration dependent effect. This change in the sugar contents revealed the disturbance in the fungus cell, which reflects the fungicidal effect of the tested compounds.

From the mentioned data, it could be concluded that 2-phenylindole and 1-acetylindole-3-butyric acid affected both RNA and DNA contents in the tested fungi, which may develop deformed and dead cells. These effects of indole acetic acid and some derivatives may due to oxidative decarboxylation leading to formation of 3-methylene-2-oxindole, which may conjugate with DNA bases and protein thiols (Folkes and Wardman, 2001). There were highly effective against polyphenoloxidase and peroxidase activities with either inhibition or elevation that means disturbance in the cell physiology as Yu and Zheng (2007) revealed that IAA alone or with *C. laurentii* stimulated catalase, peroxidase and polyphenoloxidase activities of pear fruit.. The studied indole derivatives may affect the treated fungi in another site of action as for example Kappas (1983) found that indole-3-acetic acid (IAA), indole-3-butyric acid (IBA) greatly increase somatic segregation in *Aspergillus nidulans* and increasing their concentrations increased mitotic segregation of the fungus.

Generally, from the previous mentioned results, it could be concluded that, the indole acetic acid and its derivatives can be used as good antifungal compounds.

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Table (1): melting point, NMR and Mass spectra of the prepared compounds

Compound	M. P. (^{o}C)	NMR spectrum *	Ms spectrum (m/z) **
1-Benzoylindole-3-acetic acid	117-118.5	δ: **7.45** (1H, s, 2-H), **7.47** and **7.58** (2H, d, 5-H and 6-H, *J* = 5.1 and 5.1 Hz), **7.62** (2H, d, 4-H and 7-H, *J* = 5.1 Hz), **3.85** (2H, s, CH_2), benzoyl protons at δ: **7.94** (2H, s, *m*-2H), **7.97** (1H, s, *p*-1H), **7.98** (2H, s, *o*-2H), **12.85** (1H, s, COOH)	281 (5.2) $(M+2)^+$, 207 (16.2), 193 (20.0), 175 (27.3), 130 (100), 122 (35.4), 117 (15.1), 105 (45.9), 77 (36.9), 69 (27.6), 57 (30.4)
1-acetylindole-3-butyric acid	165-166	**2.49** (3H, s, $COCH_3$), **7.05** (1H, d, 2-H, *J* = 1.0 Hz), **7.32** (2H, dd, 5-H and 6-H, *J* = 8.1 and 8.1 Hz) and **7.65** (2H, d, 4-H and 7-H, *J* = 9.0 Hz), aliphatic signals at δ: **2.94** (2H, t, 1-2H), **2.50** (2H, m, 2-2H), **2.15** (2H, t, 3-2H), **11.55** (1H, s, COOH),	223 (7.16), 193 (28.8), 185 (70.6), 157 (16.3), 143 (24.1), 129 (100.0), 128 (21.5), 97 (33.4), 83 (50.1), 69 (55.7), 55 (70.1)
1-benzoylindole-3-butyric acid	113-114	**7.45** (1H, s, 2-H), **7.48** and **7.55** (2H, d, 5-H and 6-H, *J* = 5.1 and 5.1 Hz), **7.60** (2H, d, 4-H and 7-H, *J* = 5.1 Hz), aliphatic signals at δ: **4.45** (2H, t, 1-2H), **4.05** (2H, m, 2-2H), **2.3** (2H, t, 3-2H), benzoyl protons at δ: **7.95** (2H, s, *m*-2H), **7.97** (1H, s, *p*-1H), **7.99** (2H, s, *o*-2H) **12.9** (1H, s, COOH)	306 (0.1) $(M-1^+)$, 130 (2.6), 123 (8.8), 122 (100.0), 105 (70.8), 94 (1.6), 77 (22.2), 51 (19.1)
2-Phenylindole	189-190	δ: **6.9** (1H, s, 3-H), **7.89** (2H, d, 4-H and 7-H, *J* = 8.67 Hz), **7.48** and **7.57** (2H, dd,5-H and 6-H, *J* = 8.67 and 8.67 Hz), **11.55** (1H, s, N-H), phenyl signals at **7.03** (2H, dd, *o*-2H, *J* = 8.3 Hz), **7.15** (1H, d, *p*-1H, *J* = 8.3 Hz), **7.31** (2H, d, *m*-2H, *J* = 8.3 Hz)	194 (16.8) $(M+1^+)$, 193 (100.0) (M^+), 192 (14.72) $(M-1^+)$, 165 (77.04), 89 (16.10), 83 (8.0), 63 (9.35)
1-acetyl-2-phenylindole	179-180.5	δ: **6.8** (1H, s, 3-H), **7.47** and **7.54** (2H, dd,5-H and 6-H, J = 8.67 and 8.67 Hz), **7.87** (2H, d, 4-H and 7-H, J = 8.67 Hz), phenyl signals at **7.02** (2H, dd, *o*-2H, *J* = 8.3 Hz), **7.11** (1H, d, *p*-1H, *J* = 8.3 Hz), **7.3** (2H, d, *m*-2H, *J* = 8.3 Hz) , **3.37** (3H, s, COCH3)	236 (0.16) $(M+1^+)$, 235 (0.50) (M^+), 194 (15.8), 193 (100.0), 192 (15.6), 165 (32.5), 115 (11.0), 103 (8.54), 102 (21.28), 89 (43.8), 77 (25.6), 63 (53.56)
1-benzoyl-2-phenylindole	106-107	δ: **6.89** (1H, s, 3-H), **7.96** and **7.86** (2H, dd,5-H and 6-H, *J* = 8.67 and 8.67 Hz), **7.97** (2H, d, 4-H and 7-H, *J* = 5.33 and 1.67 Hz), 2-phenyl signals at **7.02** (2H, dd, *o*-2H, *J* = 7.6, 1.0 Hz), **7.1** (1H, d, *p*-1H, *J* = 8.0 Hz) and 7.31 δ (2H, d, *m*-2H, *J* = 7.66 Hz), N-benzoyl signals at **7.43** (2H, dd, *m*-2H, *J* = 5.33 and 3.66 Hz), **7.48** (1H, dd, *p*-H, *J* = 8.3 and 2.7 Hz), **7.43** (2H, dd, *o*-2H, *J* = 8.3 Hz)	286 (27.4), 285 (20.8), 236 (19. 6), 193 (100.0), 122 (12.2), 105 (22.9), 97 (43.1), 83 (36.1), 55 (35.8)

* s: singlet; d: doublet; dd: double doublet; t: triplet; *o*-: ortho; *m*-: meta; *p*: para

** ( ) Abundance %

Table (2): *In vitro* fungicidal activity screening against the tested fungi

Fungus	Treatment	IC_{50} (95%C L) µg/ml	Slope ± S.E	χ^2	TF
F. calmorum	Indole-3-acetic acid [d*]	420 (222 – 823)	0.6 ± 0.005	4.75	2.19
	1-Benzoyl indole-3-acetic acid [c]	523 (322 – 859)	0.87 ± 0.011	7.04	2.72
	Indole-3-butyric acid [a]	576 (388 – 858)	1.28 ± 0.025	2.78	3.00
	1-Acetyl indole-3-butyric acid [i]	26.6 (21.3 –33.3)	1.41 ± 0.01	4.54	0.14
	1-Benzoyl indole-3-butyric acid [b]	513 (335 – 793)	1.03 ± 0.015	1.29	2.67
	2-Phenylindole [h]	67.4 (53.0- 85.8)	1.11 ± 0.008	8.57	0.35
	1-Acetyl-2-phenylindole [g]	86.7 (66.4 – 113)	0.98 ± 0.007	5.33	0.45
	1-Benzoyl-2-phenylindole [f]	99.9 (77 – 129.9)	1.02 ± 0.008	0.63	0.52
	Metalaxyl [e]	192 (126 – 296)	0.69 ± 0.006	3.6	1.0
M. phaseoli	Indole-3-acetic acid [a]	807 (440 – 1514)	0.81 ± 0.011	2.83	4.66
	1-Benzoyl indole-3-acetic acid [d]	572 (359 – 923)	0.97 ± 0.014	2.99	3.30
	Indole-3-butyric acid [b]	699 (458 – 1073)	1.38 ± 0.003	1.28	4.03
	1-Acetyl indole-3-butyric acid [f]	59.0 (47.0 – 74)	1.21 ± 0.009	3.87	0.34
	1-Benzoyl indole-3-butyric acid [c]	448 (325 – 622)	1.38 ± 0.026	2.62	2.59
	2-Phenylindole [i]	96 (74.6- 123.4)	1.06 ± 0.008	8.1	0.55
	1-Acetyl-2-phenylindole [h]	93 (71.7 – 120.0)	1.03 ± 0.008	7.78	0.54
	1-Benzoyl-2-phenylindole [g]	355 (247 – 514)	1.02 ± 0.001	2.18	2.05
	Metalaxyl [e]	173 (127 –237.6)	0.93 ± 0.008	4.78	1.00
P. debarianum	Indole-3-acetic acid [b]	301 (207.9- 438)	0.93 ± 0.001	3.76	1.43
	1-Benzoyl indole-3-acetic acid [b]	171 (125 – 236)	0.91 ± 0.008	3.96	0.81
	Indole-3-butyric acid [d]	249 (179 – 349)	0.98 ± 0.001	9.33	1.18
	1-Acetyl indole-3-butyric acid [g]	19 (14.4 – 24.8)	1.1 ± 0.006	1.72	0.09
	1-Benzoyl indole-3-butyric acid [a]	488.4 (319 –753)	1.0 ± 0.14	0.46	2.31
	2-Phenylindole [f]	17.7 (11.8 –26.4)	0.67± 0..004	0.48	08
	1-Acetyl-2-phenylindole [g]	15.0 (9.5 – 23.2)	0.61 ± 0.004	2.15	0.07
	1-Benzoyl-2-phenylindole [e]	81 (57.2 – 115)	0.73 ±0.005	3.99	0.38
	Metalaxyl [c]	211 (145 – 310)	0.80 ± 0.007	1.03	1.00
R. solani	Indole-3-acetic acid [c]	1009 (539–1923)	0.94 ±0.016	4.42	4.36
	1-Benzoyl indole-3-acetic acid [c]	1244 (633–2515)	0.71 ± 0.008	9.08	5.38
	Indole-3-butyric acid [a]	644 (368 – 1151)	0.79 ± 0.01	2.38	2.78
	1-Acetyl indole-3-butyric acid [i]	117 (97.6 – 141)	1.57 ± 0.018	6.1	0.51
	1-Benzoyl indole-3-butyric acid [b]	663 (377 – 1192)	0.79 ± 0.01	2.64	2.87
	2-Phenylindole [h]	34.6 (25.1 –47.5)	0.81 ± 0.005	7.14	0.15
	1-Acetyl-2-phenylindole [f]	37.5 (27.6 –50.7)	0.85 ± 0.005	1.79	0.16
	1-Benzoyl-2-phenylindole [d]	122.2 (93 –161)	1.0 ±0.008	3.31	0.53
	Metalaxyl [e]	231 (167.7 –321)	0.98 ± 0.01	3.21	1.00

TF: Toxicity factor related to Metalaxyl * $p > 0.05$ against each fungus Degree of freedom = 4

Table (3): *In vivo* effect of 2-phenylindole and 1-acetylindole-3-butyric acid on polyphenoloxidase activity

Compound	Fungus	IC_{50} (µg/ml) (95%C L)	Slope ± S.E	χ^2	DF
2-phenylindole	*F. calmorum*	87.6 (68.1 – 120.7)	1.31 ± 0.02	1.87	3
	M. phaseoli	117.1 (77.8 – 205.4)	0.75 ± 0.03	0.91	3
	P. debarianum *	35.1 (25.0 – 56.6)	1.14 ± 0.02	0.54	3
	R.. solani	80.3 (56.7-131.5)	1.23 ± 0.026	0.64	3
1-acetylindole-3-butyric acid	*F. calmorum*	41.5 (29.3 – 71.8)	0.90 ± 0.019	2.18	3
	M. phaseoli	80.2 (49.6 – 165.2)	0.65 ± 0.017	1.41	3
	P. debarianum	45.6 (30.4 – 81.7)	1.03 ± 0.022	2.55	3
	R.. solani	-	-	-	-

* The enzyme activity was increased at all concentrations

Table (4): Effect of 2-phenylindole and 1-acetylindole-3-butyric acid on peroxidase activity

Compound	Fungus	Tested concentrations (of IC_{50}**)					IC_{50}	Slope ± S.E	χ^2
		0.1	0.25	0.5	1.0	2.0			
2-Phenylindole	*F. calmorum*	7.5± 0.87	26.2± 1.31	53.8± 0.76	60.1± 0.17	69.2± 0.91	49.9 (41.1-61.3)	1.45± 0.02	2.6
	M. phaseoli	- 39± 0.87	-32.2± 0.35	26.7± 0.76	40.7± 1.15	54.3± 0.58	-		
	P. debarianum	19.3± 1.53	3.7± 0.64	0.0± 0.0	-11.9± 0.9	-22.0± 0.50	-	-	-
	R.. solani *	0.0± 0.0	-46.3± 1.53	-56.3± 2.51	-67.7± 0.31	-151± 2.65	14.5 (12.8- 16.3)	2.81 ± 0.04	5.8
1-Acetylindole-3-butyric acid	*F. calmorum*	85± 0.76	23.7± 1.53	-7.9± 0.26	-23.1± 1.01	-115± 3.06	-	-	-
	M. phaseoli *	-66.7± 2.31	-111± 3.51	-139± 2.52	-150± 1.53	-261± 5.77	< 5.9	-	-
	P. debarianum	0.0± 0.0	3.0± 0.1	1.3± 0.15	19.1± 1.01	25.7± 1.15	96.14 (59.0 - 226)	1.34 ± 0.045	4.4
	R.. solani *	-275± 5.29	-212± 2.64	-175± 4.58	-119± 1.53	-87.7± 2.88	< 11.7	-	-

*The enzyme activity was increased at all concentrations ** IC_{50} in µg/ml (Table 1)

Table (5): Effect of 2-phenylindole on DNA and RNA contents

Fungus	* Tested Concentrations in µg/ml (rates of its IC_{50} values)											
	RNA content (mg/L)						DNA content (mg/L)					
	0	0.1	0.25	0.5	1.0	2.0	0	0.1	0.25	0.5	1.0	2.0
F. calmorum [c]	51.5 ±1.5	29.7 ±0.9	24.3 ±0.74	18.7 ±0.65	26.3 ±0.6	24.7 ±0.6	49.5 ±1.6	28.6 ±0.9	23.7 ± 0.7	18.0 ±0.5	25.3 ±0.6	23.8 ±0.7
M. phaseoli [a]	16.1 ±0.4	6.7 ± 0.1	7.2 ±0.09	8.3 ±0.65	6.3 ±0.1	5.9 ±0.1	15.5 ±0.5	6.4 ±0.04	6.8 ±0.03	8.0 ±0.1	6.1 ±0.1	5.7 ±0.03
P. debarianum [b]	32.2 ±0.9	32.1 ± 0.7	19.8 ±0.8	11.4 ±0.8	30.6 ±1.2	24.2 ±0.9	31..0 ±0.8	30.9 ±0.9	19.1 ± 0.78	10.9 ±0.7	29.4 ±0.6	23.3 ±0.8
R. solani [c]	25.0 ±0.7	29.4 ± 0.8	30.0 ±1.0	30.8 ±1.1	32.2 ±1.2	28.0 ±1.1	24.0 ±0.7	28.3 ±0.75	28.9 ± 0.8	29.6 ±0.9	30.9 ±0.7	27.0 ±0.8

LSD_{RNA} Conc. = 0.4104 LSD_{DNA} Conc = 0.4071 * IC_{50} in µg/ml (Table 1)
LSD_{RNA} Fung. = 0.3351 LSD_{DNA} Fung = 0.3324

Table (6): Effect of 1-acetylindole-3-butyric acid on DNA and RNA contents

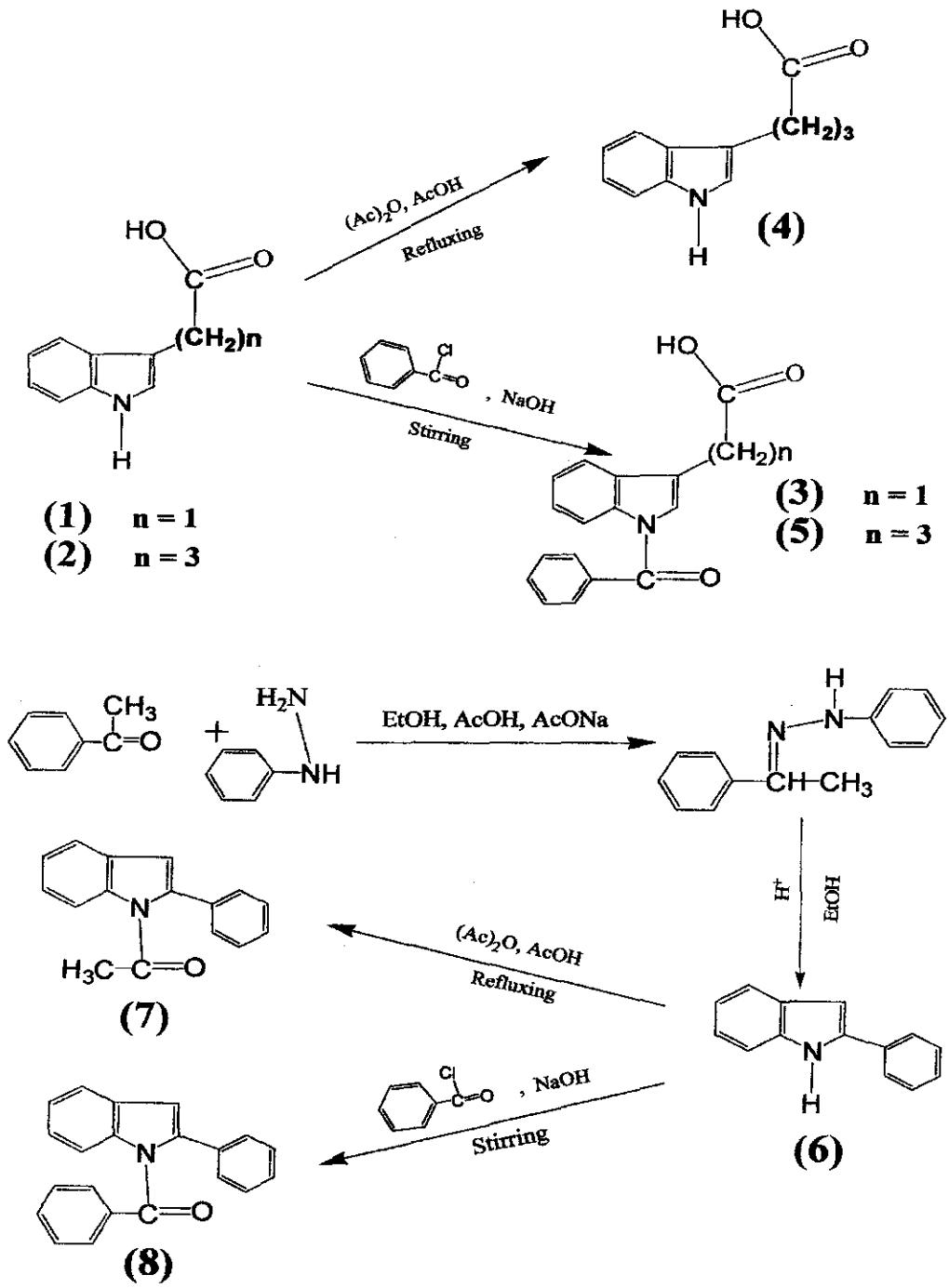
Fungus	* Tested Concentrations in µg/ml (rates of its IC_{50} values)											
	RNA content (mg/L)						RNA content (mg/L)					
	0	0.1	0.25	0.5	1.0	2.0	0	0.1	0.25	0.5	1.0	2.0
F. calmorum [d]	51.5 ±1.5	45.4 ±1.3	31.1 ±0.5	29.2 ±0.4	28.3 ±0.9	20.3 ±0.4	49.5 ±1.6	43.6 ±1.2	29.9 ±1.2	28.0 ±0.8	27.2 ±0.6	19.5 ±0.6
M. phaseoli [a]	16.1 ±0.4	12.9 ±0.5	10.6 ±0.1	8.6 ±0.1	8.5 ±0.3	6.7 ±0.2	15.5 ±0.5	11.7 ±0.9	10.3 ±0.3	8.3 ±0.3	8.2 ±0.2	6.5 ±0.3
P. debarianum [b]	32.2 ±0.9	27.0 ±0.9	26.8 ±0.7	11.8 ±0.4	18.2 ±0.6	22.5 ±0.6	31..0 ±0.8	26.0 ±0.8	25.8 ±1.1	11.3 ±0.4	17.5 ±0.4	21.6 ±0.9
R. solani [c]	25.0 ±0.7	23.6 ±0.7	26.2 ±0.8	27.9 ±0.55	32.6 ±1.0	36.1 ±0.8	24.0 ±0.7	23.2 ±0.7	25.2 ±1.0	26.6 ±0.8	31.4 ±0.9	34.7 ±0.9

LSD_{RNA} Conc. = 0.5471 LSD_{DNA} Conc = 0.6687 * IC_{50} in µg/ml (Table 1)
LSD_{RNA} Fung. = 0.4467 LSD_{DNA} Fung = 0.5460

Table (7): Effect of 2-phenylindole and 1-acetylindole-3-butyric acid on *M. phaseoli* and *R. solani* sugar contents

Compound	Fungus	Sugar type	Sugar contents (mg/gm dry weight) at different rates of its IC_{50} values				
			0	0.1 f *	0.25 f	0.5 f	1.0 f
2-phenylindole	*M. phaseoli*	R.S	7.3±0.5	6.3±0.4	4.6±0.1	2.9±0..1	5.8±0.3
		Non-R.S	12.0±0.8	2.4±0.1	5.1±0.1	1.4± 0.2	1.3±0.1
		T.S.S	19.3±0.75	8.7±0.3	9.7±0.3	4.3±0.3	7.1±0.5
	R. solani	R.S	17.3±1.0	44.3±1.2	60.3±1.2	16.0±0.5	7.7±0.4
		Non-R.S	2.8±0.2	14.1±0.6	11.4±0.4	3.1±0.2	2.2±0.2
		T.S.S	20.1±0.8	58.4±1.4	71.8±1.5	19.1±0.8	9.8±0.4
1-Acetylindole-3-butyric acid	*M. phaseoli*	R.S	7.3±0.5	2.5±0.1	14.8±0.4	20.8±1.1	25.8±1.0
		Non-R.S	12.0±0.8	1.7±0.04	7.4±0.2	10.2±0.5	8.9±0.4
		T.S.S	19.3±0.75	4.2± 0.2	22.2±0.7	31.0±1.2	34.7±0.9
	R. solani	R.S	17.3±1.0	9.7±0.6	9.6±0.3	5.6±0.4	2.6±0.2
		Non-R.S	2.8±0.2	2.5±0.2	1.2±0.1	0.4±0.1	0.2±0.1
		T.S.S	20.1± 0.8	12.2±0.4	10.8±0.4	6.0±0.5	2.8±0.3

Data are averages of three replicates. * f is IC_{50} value from Table (1)
R.S: reduced sugars; **Non- R.S:** non reducing sugars; **T.S.S:** total soluble sugars



1	Indole-3-acetic acid	5	1-Benzoyl indole-3-butyric acid
2	Indole-3-butyric acid	6	2-Phenylindole
3	1-Benzoyl indole-3-acetic acid	7	1-Acetyl-2-phenylindole
4	1-Acetyl indole-3-butyric acid	8	1-Benzoyl-2-phenylindole

Fig. 1: Preparation scheme of compounds 1-8

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الملخص العربى

النشاط الابادى لبعض مشتقات الاندول على الفطريات

أحمد صبرى عبد العاطى

الاسكندرية- مصر- الشاطبى – جامعة الإسكندرية – كلية الزراعة -قسم كيمياء المبيدات

تم تحضير ستة من مشتقات الإندول و هى الـ ١-بنزويل اندول-٣-أسيتيك أسيد ، ١-أسيتيل اندول-٣-بيوتيريك أسيد ، ١-بنزويل اندول-٣- بيوتيريك أسيد ، ٢-فينيل اندول ، ١-أسيتيل- ٢-فينيل اندول ، ١-بنزويل-٢-فينيل اندول كمشتقات للأكسينات الطبيعية. تم التعرف على هذه المركبات طيفيا باستخدام الرنين النووى المغناطيسى NMR و مطياف الكتلة Mass spectroscopy.

تم تقدير النشاط الإبادى لهذه المركبات إضافة إلى الـ إندول-٣-أسيتيك أسيد (IAA) ، إندول-٣- بيوتيريك أسيد (IBA) على مجموعة من الفطريات الهامة جدا إقتصاديا و هى الـ فيوزاريوم كالمورام ، بيثيوم ديباريانم ، رايزوكتونيا سولانى و كذلك فطر الـ ماكروفوميتا فاصيولاى مقارنة بمركب الـ Metalaxyl (Radomil) الذى أستخدم كمبيد فطرى قياسى.

ظهر التأثير الابادى لهذه المركبات كدالة فى كل من الفطر و المركب المطبق عليه. تم تقدير التركيز المؤثر على ٥٠% من النمو الهيفى للفطر لكل من هذه المركبات على كل فطر و التوصل للعلاقة بين التركيب الكيماوى و التأثير الحيوى لهذه المركبات على الفطريات المختبرة.

تم دراسة تأثير كل من مركبى الـ٢-فينيل إندول ، الـ ١-أسيتايل إندول-٣-بيوتيريك أسيد كمثال للمركبات الفعالة على كل من انزيمى البولى فينول أكسيديز و البيروكسيديز داخليا *In Vivo* . كما تم أيضا دراسة تأثيرهما على محتوى الـ RNA and DNA contents بالإضافة إلى محتوى السكريات فى الفطريات محل الدراسة. وجد أن انزيم البولى فينول أكسيديز قد تأثر طرديا بتركيز مركب الـ ٢- فينيل إندول بتركيز مثبط لـ ٥٠% من نشاط الانزيم قدره ٨٠,٣ ميكروجرام/ مل. تعدى مركب الـ ١-أسيتايل إندول-٣-بيوتيريك أسيد مركب الـ ٢-فينيل إندول فى تأثيره على هذا الأنزيم بتركيز مثبط لـ ٥٠% قدره ٤١,٥ و ٨٠,٢ مقارنة بـ ٨٧,٦ و ١١٧,١ ميكروجرام/ مل على الترتيب فى حالة فطريات الـ*F. calmorum* and *M. phaseoli*. بينما فى حالة الـ *P. debarianum* ثبط الأنزيم بتركيز مثبط لـ ٥٠% قدره ٤٥,٦ ميكروجرام/ مل ، تم تنشيطه على كل تركيزات المركب الأول. وجد أن انزيم البيروكسيديز تأثر بصورة إختلفت بإختلاف الفطر المعامل. قد يعزى هذا الإختلاف فى التأثير على الإنزيمات إلى نوع المستبدل و موضع استبداله على حلقة الاندول نفسها.

تعدى مركب الـ ٢-فينيل إندول فى تأثيره مركب الـ ١-أسيتايل إندول-٣-بيوتيريك أسيد على الـ RNA and DNA contents فى فطريات الـ *F. calmorum, M. phaseoli* and *R. solani* ، فى حين تقريبا تعادل تأثيرهما على محتوى فطر الـ *P. debarianum* متوقفا على تركيز المركبز. تأثر محتوى الفطريات المعاملة من السكريات مفسرا حدوث خلل فيسيولوجى بالخلية وكذلك إحداث تغيير لتركيب الخلايا بإنتاج خلايا غير كاملة النمو مشوهة منتهيا الى موتها.