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# N-САЛИЦИЛОИЛАМИДЫ И ИХ СОЛИ: СИНТЕЗ, БИОЛОГИЧЕСКАЯ АКТИВНОСТЬ И ТОКСИЧНОСТЬ

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***N*-салицилоилпроизводные морфолина, глицина и гамма-аминомасляной кислоты были синтезированы по реакции Шоттена-Баумана путем взаимодействия салицилоилхлорида с морфолином или глицином с высоким выходом. Натриевая и литиевая соли были получены путем взаимодействия салициламидов с этилатом натрия или гидроксидом лития в инертном растворителе. Для определения качественного и количественного состава реакционных масс были использованы следующие методы анализа: тонкослойная хроматография, спектроскопия ядерного магнитного резонанса (ЯМР-спектроскопия) и элементный анализ. Количественный анализ ионов металлов проводили потенциометрическим методом. Также был проведен прогноз потенциальной биологической активности синтезированных соединений с помощью анализа связей структура-активность в программе PASS Online. У синтезированных солей с высокой вероятностью (Pa > 0,8) была выявлена психотропная активность и с вероятностью Pa около 0,5 противовирусная. Для выявления соединений-лидеров психотропная («открытое поле», приподнятый «плюс-лабиринт», метод условной реакции пассивного избегания, тест «принудительного» плавания Порсольта), анальгетическая (измерение порога вокализации) активности, а также острая токсичность оценивались в эксперименте. Полученные данные показали, что динатриевая соль *N*-салицилоилглицина (салицилурат динатрия) проявляет выраженное антиамнестическое действие, дилитиевая соль *N*-салицилоилглицина (салицилурат дилития) и натриевая соль салицилоилморфлида показали значительную антидепрессивную активность и антиамнестическое действие, литиевая соль салицилоилморфлида – психостимулирующее действие, гамма-*N*-салицилоиламинобутират лития – психостимулирующую и антиамнестическую активности. Исследования острой токсичности LD50 показали, что синтезированные соли являются малотоксичными. Была изучена противовирусная активность в отношении большого количества ДНК- и РНК-вирусов, включая вирусы простого герпеса 1 и 2 типов, вирус оспы, вирус везикулярного стоматита, ВИЧ-1 и ВИЧ-2. Биологическая активность соединений также оценивалась в отношении клеток карциномы шейки матки человека (HeLa) и Т-лимфоцитов (CEM), а также клеток лейкемии (L1210). Исследования противовирусной активности показали, что ни одно из протестированных соединений не было активным против ДНК- или РНК-вирусов.***

**Ключевые слова:** салицилоиламиды, глицин, морфолин, ГАМК, соли щелочных металлов, биологическая активность, токсичность

# N-SALICYLOYL AMIDES AND THEIR SALTS: SYNTHESIS, BIOLOGICAL ACTIVITY AND TOXICITY

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***The Schotten-Baumann reaction was used to synthesize N-salicyloyl derivatives of Morpholine, Glycine and gamma-aminobutyric acid in the process between salicyloyl chloride with morpholine, glycine or gamma-aminobutyric acid with high yield. Sodium and lithium salts of the salicylamides were synthesized by the reaction of an amide with sodium ethylate or lithium hydroxide in inert solvent. The synthesized compounds were charachterized by $^1$H nuclear magnetic resonance spectra and elemental analysis. Purity was checked by thin layer chromatography. Quantitative analysis of metal ions was done by potentiometry. To select the most promising compounds exhibiting psychotropic (Pa > 0.8) and antiviral (Pa about 0.5) activities, computer analysis was carried out through the PASS program. In the present study, psychotropic (the open-field exploratory test, the forced swim test, the elevated plus-maze test, the passive avoidance test), analgesic (current vocalization threshold) activities and acute toxicity were evaluated. The results of psychotropic activity revealed that amides possess significant psychotropic activity and low acute toxicity. Disodium salicylurate showed a significant antiamnesic action. The results suggested a significant antidepressant activity and antiamnesic action for dilithium salicylurate and sodium salt of salycyloyl morphlide, a psychostimulant action for lithium salt of salicyloyl morphlide. Lithium gamma-(N-salycylamino)butyrate demonstrated psychostimulant and antiamnesic activities. Our present investigation demonstrated that all compounds are very safe for consumption with high LD50 value. All obtained compounds were evaluated for their antiviral activities against a large number of DNA and RNA viruses including herpes simplex viruses 1 and 2, vaccinia virus, vesicular stomatitis virus, HIV-1 and HIV-2. These compounds were evaluated against human cervix carcinoma cells (HeLa) and CEM T-lymphocytes as well as murine leukemia cells (L1210). The antiviral activity studies depicted that none of the tested compounds were active against DNA or RNA viruses.***

**Key words:** salicyloyl amides, Glycine, Morpholine, gamma-aminobutyric acid (GABA), alkali metals salts, biological activity, toxicity

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## INTRODUCTION

Derivatives of salicylic acid (SA) are known to possess a number of biological activities such as antimicrobial, antipyretic, anti-inflammatory, antithrombotic, analgesic, anticonvulsant. Salicyloyl morpholide is known as an anti-inflammatory agent and can be used in combination with substances capable of exerting an anti-inflammatory effect. Salicyloyl glycine (salicyluric acid) inhibits peptidyl α-hydroxylating monooxygenase (PHM) which functions in vivo promoting the biosynthesis of α-amidated peptide hormones in mammals and insects. PHM is a potential target for the development of inhibitors as drugs for the treatment of human diseases such as in cancer, rheumatoid arthritis, anxiety and depression. Gamma-aminobutyric acid (GABA) is a well-known neurotransmitter, a chemical messenger in the brain [1-9].

To select from them the most promising compounds exhibiting psychotropic properties, we preliminarily carried out computer analysis using the PASS program [10]. As is known, the result of the prognosticating biological activity is represented in PASS in the form of probabilities Pa (active) and Pi (inactive). If Pa > 0.7, the chance to experimentally detect any activity is quite high, and the compound is most likely to combine the most important features of active compounds and may even turn out to be a precursor of a new chemical class for the considered type of biological activity. The calculations have shown that synthesized compounds might be active in phobic disorder treatment with Pa > 0.8 and confidence antiviral activity about 0.5 [11]. In the present study, we examined the possible psychotropic activity of water soluble salts of salicyloyl morpholide, salicyluric acid and gamma-(N-salicylamino)butyric acid. We also studied the effect of a cation (Na+, Li+) on the activity. For instance, lithium has been used in the treatment of schizophrenic and schizoaffective disorders. Based on this data, it will be possible to assume the expediency of a further study of the compounds as potent phamacologically active substances.

## EXPERIMENTAL

The $^1$H NMR spectra were recorded on a Bruker Advance 300 spectrometer (USA) using DMSO-$d_6$ as solvent and tetramethylsilane as reference. Elemental analysis was performed with a Perkin Elmer 2400 Series II analyzer (USA). Water soluble forms of amides were prepared in the form of alkali metal salts. Purity of salicyloyl amides was checked by the Thin layer chromatography using silica gel G as stationary phase and ethyl acetate : hexane (3:1) as mobile phase. The spots resolved were visualized as brown colored spots by using Iodine chamber. Melting points were determined in capillaries on a Stuart SMP-30 apparatus at heating rate 3.5 °C/min.

Salicyloyl amides were synthesized from salicyloyl chloride and amino compound (Morpholine, Glycine and GABA) as given [12].

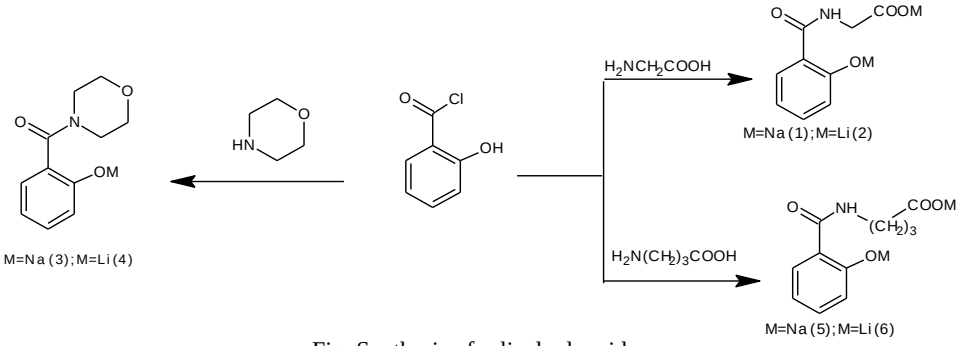


Fig. Synthesis of salicyloyl amides
Рис. Синтез салицилоиламидов

The structure of obtained compounds corresponded to literature date. The morpholine fragment was represented by two multiplets in the region at δ 3.19-3.32 ppm and 3.39-3.51 ppm.

Salts of salicylamides were synthesized by the reaction of amide with sodium ethylate or anhydrous LiOH in inert organic solvent at room temperature [13].

*Disodium Salicylurate (1)*. Yield 2.34 g (98%). Found, %: C 45.19; H 2.94; N 5.85; Na 19.21. $C_9H_7NNa_2O_4$. Calculated, %: C 45.20; H 2.95; N 5.86; Na 19.23.

*Dilithium Salicylurate (2)*. Yield 1.98 g (96%). Found, %: C 52.20; H 3.43; N 6.76; Li 6.70. $C_9H_7Li_2NO_4$. Calculated, %: C 52.21; H 3.41; N 6.77; Li 6.71.

*N-Salicyloyl morpholide sodium salt (3).* Yield 2.04 g (89%). Found, %: C 57.41; H 5.25; N 6.12; Na 10.09. $C_{11}H_{12}NNaO_3$. Calculated, %: C 57.64; H 5.28; N 6.11; Na 10.03.

*N-Salicyloyl morpholide lithium salt (4).* Yield 2.08 g (96%). Found, %: C 62.51; H 5.62; N 6.62; Li 3.29. $C_{11}H_{12}LiNO_3$. Calculated, %: C 61.98; H 5.67; N 6.57; Li 3.26.

*Disodium gamma-(N-salicyloylamino)butyrate (5).* Yield 2.58 g (97%). Found, %: C 45.45; H 5.23; N 6.13; Na 10.10. $C_{11}H_{11}NNa_2O_4$. Calculated, %: C 49.45; H 4.15; N 5.24; Na 17.21.

*Dilithium gamma-(N-salicyloylamino)butyrate (6).* Yield 2.25 g (96%). Found, %: C 56.18; H 4.73; Li 5.88; N 5.96. $C_{11}H_{11}Li_2NO_4$. Calculated, %: C 56.2; H 4.72; Li 5.90; N 5.96.

The study of psychotropic and analgesic activities was conducted at the Volgograd State Medical University. When conducting experiments on animals, the rules of laboratory practice and ethical requirements were observed. All animals were delivered from the nursery of laboratory animals "Rappolovo" [14-16].

Albino rats Wistar of either sex were weighted (180-200 gm). A total of 150 animals were divided into 3 groups (I, II, III). Group I: normal control group – received 0.9% sodium chloride solution (physiologic saline) in a constant volume of 1 ml/kg as vehicle, Group II: received the studied salt solution in dose of 10 mg/kg, Group III: received the studied salt solution in dose of 50 mg/kg. Studied compounds were prepared in 0.9% sodium chloride solution and administered intraperitoneally (i.p.).

Originally introduced as a measure of emotional behavior in rats, open field exploration has proven to be equally successful with rodents. The test provides a unique opportunity to systematically assess novel environment exploration, general locomotor activity, and provide an initial screen for anxiety-related behavior in rodents. In addition, repeated exposure or extended session length provides a method for assessing habituation to the increasingly familiar chamber environment. It has been suggested that two factors influence anxiety-like behavior in the open field. The first is social isolation resulting from the physical separation from cage mates when performing the test. The second is the stress created by the brightly lit, unprotected, novel test environment. The procedure involves

forced confrontation of a rodent with the situation. The animal is placed in the center or close to the walls of the apparatus and the following behavioral items are recorded for a period ranging from 5 to 20 min: horizontal locomotion, and frequency of rearing or leaning (vertical locomotion). In such a situation, rodents spontaneously prefer the periphery of the apparatus to activity in the central parts of the open field. Increase in time spent in the central part as well as of the ratio central/total locomotion or decrease of the latency to enter the central part are indications of anxiolysis. In the test, the apparatus consisted of a wooden box (60×60×30 cm). The base of the box was divided into 16 squares (15×15 cm). The apparatus was illuminated with a 40 W lamp suspended 100 cm above it. Animals were treated with a compound (10, 50 mg/kg) or physiologic saline. 45-60 min after administration, the animal was placed in the same corner of the field. The measurements included distance moved (horizontal locomotion), frequency of visits to the central area, number of rearing events (vertical locomotion).

Among all animal models, the forced swimming test (FST) remains one of the most used tools for screening antidepressants. Two parameters were defined in the evaluation of FST: 1) Immobility – defined in the traditional Porsolt test as when no additional activity is observed other than that required to keep animal's head above water; 2) Climbing behavior – being defined as the upward-directed movement (vertical) of the forepaws along the side of the swim chamber. Rats were housed in individual cages with free access to water in a temperature-controlled facility with a 12 h light/dark cycle. Briefly, animals were placed at day one in a large cylinder (30 cm in diameter and 45 cm deep) of 24-28 °C water for a 15-min period. At day two (24 h later), treated rats (45-60 min post intraperitoneal injection of drug in two dosages 10 and 50 mg/kg) were dropped to the cylinder for 6 min and scoring was performed in the last 4 min after 2 min adaptation. The immobility in the animals included minor movements, which are strictly necessary to maintain the animal's head above water. The criterion for antidepressant activity was considered statistically significant reductions in immobilization duration.

The elevated plus-maze is the most widely used test to measure fear or anxiety and it is particularly sensitive to anxiety-reducing drugs. The plus-maze was made of plywood and consisted of two open arms (21.5×7.5 cm) and two enclosed arms (21.5×7.5×20 cm) which extended from a central 7.5×7.5 cm platform. The plus-maze was elevated 38 cm above the floor. The enclosed arms were painted black. Anxious animals refrain from entering the open arm and prefer the closed arm. In the experiment, rodents were administered intraperitonealy with 10 and 50 mg/kg of a compound. 45-60 min later, each rat was placed in the central square of the plus-maze facing a closed arm, and its behavior was observed for 5 min. The number of entries onto open and closed arms and the times spent in open and closed arms and in the central square were scored.

In the passive (inhibitory) avoidance test the subjects learn to avoid a mild aversive stimulus by remaining in the lighted side of a two-compartment chamber and not entering the dark (unsafe) side. Observers record the median step-through latency (i.e., number of seconds elapsed before subjects cross into the unsafe side of the chamber), as well as the time spent in dark side. This test is a quick, simple method of assessing the effect of drugs on memory retention. A two-way shuttle-box with acrylic walls and steel floor bars (with diameter of 0.7 mm, and 8 mm apart) was used. The box is divided into two compartments (15×9.5×16.5 cm each one). The safe compartment is white and continuously illuminated, whereas the «shock» compartment is dark. The two compartments are separated by a partition with sliding guillotine door at floor level. In the experiment, animals were injected studied compounds or saline immediately after the training phase. Animals were randomly subjected to inhibitory avoidance task. Training and test phases began with a 90-s adaptation period to the apparatus in the light compartment. Following this, the door between the compartments was opened and time taken to enter the dark compartment, defined as latency. In the training phase, the door was opened and the animal was allowed to stay in the light was closed and a foot-shock (0.5 mA for 5 s) was delivered through the grid floor compartment for a maximum of 300 s. As soon as the animal entered the dark compartment the sliding door.

Analgesic activity was determined on rats of both sexes. Rats were acclimatized to the laboratory for 1 week prior to the experiment. All the experimental animals had free access to water and food pellets. The animals were fasted overnight before use. On day of experiment non fasted animals were weighed and dosed as per randomization, test compounds were dosed intraperitonealy before one hour of the pain induced electrically. In the test, the apparatus consisted of a plastic box with electrode floor. The animal was placed in the box and electrical impulses were given with timed voltage ramp (velocity of voltage ramp – 5 V/s, impulse duration – 0.2 s). Supramaximal intensity of impulse was as high as the animals could tolerate. The vocalization threshold was recorded.

All experimental results were processed statistically with the calculation of the Fisher-Student criterion [17, 18]. The effect was considered reliable at $p < 0.05$.

White mice, weighting 25 to 30 mg, were used for acute toxicity study [19]. The test material was administered intraperitoneally at the dosage levels from 400 up to 2000 mg/kg. Acute toxicity studies involved two parts: (1) a range finding study to establish the tolerated dose range and (2) an acute toxicity study to establish the maximum tolerated dose. Observations for pharmacotoxic signs and mortality were made at 24 h and daily thereafter for a total of 15 days. Body weights were recorded prior to fasting, immediately preceding dosing and at 15 days.

Antiviral activity studied by Rega Institute for Medical Research (Belgium), Laboratory of virology and chemotherapy. When conducting experiments on animals, the rules of laboratory practice were followed [20, 21]. Inhibition of HIV-1(IIIB)- and HIV-2(ROD)-induced cytopathicity in CEM cell cultures was measured in microtiter 96-well plates containing ~ $3 \cdot 10^5$ CEM cells/mL infected with 100 $CCID_{50}$ (1 $CCID_{50}$ being the virus dose to infect 50% of the cell cultures) of HIV per milliliter and containing appropriate dilutions of the test compounds.

***Table 1***

**Effect of synthesized compounds in open-field exploratory test, in forced swim test and antianxiety activity in the elevated plus-maze test**

***Таблица 1.*** **Эффект синтезированных соединений в исследовательском тесте «открытое поле», в тесте принудительного плавания и противотревожная активность в тесте «приподнятый крестообразный лабиринт»**

| Compound | Dose, mg/kg | Vertical locomotion, s | Horizontal Locomotion, s | Frequency of visits to the central area | Immobility, s | Climbing behavior, s | Frequency of entries to the open arms | Time spent in the open arms, s |
|---|---|---|---|---|---|---|---|---|
| Control | 0 | 28.88±7.11 | 3.25±0.68 | 1.00±0.50 | 107.26±26.48 | 0.50±0.25 | 1.38±0.17 | 38.5±14.08 |
| **1** | 10 | 13.38±3.52 | 1.75±0.79 | 6.00±0.83[a] | 99.88±26.14 | 0.38±0.35 | 1.13±0.28 | 22.13±10.44 |
| | 50 | 21.38±0.35 | 3.50±1.02 | 3.63±0.95 | 151.88±19.36 | 0.00 ± 0.00 | 1.63±0.25 | 30.88±9.16 |
| Control | 0 | 3.75±0.91 | 15.63±7.57 | 1.38±0.68 | 79.50±17.10 | 0.00 ± 0.00 | 1.25±0.34 | 25.88±10.71 |
| **2** | 10 | 5.75±2.16 | 34.50±6.51 | 0.75±0.34 | 35.50±7.59[a] | 0.00±0.00 | 2.38±0.47 | 28.25±5.82 |
| | 50 | 7.50±2.19 | 38.00±8.20 | 0.63±0.17 | 37.38±5.60 | 1.50±0.50[a] | 2.50±0.31 | 44.63±10.35 |
| Control | 0 | 7.00±0.83 | 25.38±4.67 | 0.38±0.17 | 115.75±13.21 | 0.63±0.30 | 1.25±0.49 | 21.25±8.88 |
| **3** | 10 | 8.75±1.74 | 35.25±6.09 | 0.38±0.17 | 86.88±16.32 | 0.25±0.15 | 1.14±0.31 | 16.00±5.49 |
| | 50 | 14.43±1.05[a] | 48.43±6.83[a] | 0.57±0.34 | 100.71±27.02 | 0.71±0.44 | 2.86±0.24[a] | 31.29±5.39[a] |
| Control | 0 | 3.75±0.91 | 15.63±7.57 | 1.38±0.68 | 79.50±17.10 | 0.00±0.00 | 1.25±0.34 | 25.88±10.71 |
| **4** | 10 | 5.00±1.71 | 23.38±5.33 | 0.38±0.25 | 52.63±15.40 | 0.75±0.34 | 1.75±0.29 | 37.63±7.91 |
| | 50 | 9.50±2.34 | 33.13±4.43[a] | 0.25±0.15 | 17.00±5.44[a] | 1.13±0.41 | 2.25±0.42 | 33.38±4.79 |
| Control | 0 | 23.28±6.43 | 9.50±2.39 | 1.00±0.59 | 112.88±7.42 | 0.25±0.23 | 1.00±0.25 | 43.25±14.46 |
| **5** | 10 | 23.75±6.18 | 5.88±1.24 | 0.00±0.31 | 136.25±12.39 | 0.25±0.23 | 1.25±0.29 | 46.38±11.79 |
| | 50 | 26.50±3.58 | 4.38±0.61 | 0.88±0.41 | 118.13±16.09 | 0.25±0.23 | 1.13±0.33 | 44.63±17.35 |
| Control | 0 | 3.75±0.91 | 15.63±7.57 | 1.38±0.68 | 79.50±17.10 | 0.00±0.00 | 1.25±0.34 | 25.88±10.71 |
| **6** | 10 | 6.88±1.28 | 46.38±10.85[a] | 0.75±0.29 | 29.13±9.27[a] | 1.50±0.50* | 1.88±0.37 | 24.75±4.03 |
| | 50 | 1.13±0.54 | 14.13±3.02 | 0.13±0.12 | 33.50±8.17[a] | 0.13±0.12 | 1.88±0.33 | 38.88±8.69 |

Note: [a] statistical significance $p<0.05$ versus control group. Data were expressed as mean ± SEM

Примечание: [a] статистическая значимость $p<0,05$ по сравнению с контрольной группой. Данные выражали как среднее ± SEM

***Table 2***

**Effect of synthesized compounds in the passive avoidance test and analgesic activity. Acute toxicity of synthesized compounds**

***Таблица 2.*** **Влияние синтезированных соединений в тесте пассивного избегания и анальгетическая активность. Острая токсичность синтезированных соединений**

| Compound | Dose, mg/kg | Latency, s | Time spent in dark side, s | vocalization threshold, V | $LD_{50}$, mg/kg |
|---|---|---|---|---|---|
| 1 | 2 | 3 | 4 | 5 | 6 |
| Control | 0 | 44.63±14.30 | 136.63±14.71 | 45.63±3.69 | |
| **1** | 10 | 119.33±30.11 | 60.67±33.11[a] | 38.38±1.51 | 1729.56 |
| | 50 | 80.57±21.51 | 102.29±22.09 | 35.63±3.85 | |
| Control | 0 | 19.13±3.20 | 160.88±3.20 | 32.57±2.17 | |
| **2** | 10 | 152.43±23.10[a] | 27.57±23.10[a] | 39.38±3.05 | 1233.92 |
| | 50 | 143.00±22.86[a] | 37.00±22.86[a] | 42.00±3.76 | |
| Control | 0 | 19.13±3.20 | 160.88±3.20 | 32.57±2.17 | |
| **3** | 10 | 108.00±31.44[a] | 72.00±31.44[a] | 28.57±3.01 | 1346.43 |
| | 50 | 35.00±15.20 | 145.00±15.20 | 61.43±5.88[a] | |

| | | | | | |
|---|---|---|---|---|---|
| Control | 0 | 19.13±3.20 | 160.88±3.20 | 32.57±2.17 | |
| **4** | 10 | 131.29±23.48 | 48.71±23.40[a] | 31.63±2.83 | 1473.62 |
| | 50 | 180.0±00.00[a] | 0.00±0.00[a] | 41.88±3.13[a] | |
| Control | 0 | 45.60±30.14 | 134.40±30.14 | 35.75±3.13 | |
| **5** | 10 | 110.13± 5.59 | 69.88±25.59 | 42.13±3.57 | 1009.35 |
| | 50 | 63.71±27.97 | 116.29±27.97 | 39.38±3.92 | |
| 1 | 2 | 3 | 4 | 5 | 6 |
| Control | 0 | 19.13±3.20 | 160.88±3.20 | 32.59±2.17 | |
| **6** | 10 | 85.29±26.74 | 94.71±26.74 | 41.50±4.34 | 1346.43 |
| | 50 | 180.0±0.00[a] | 0.00±0.00[a] | 31.88±2.10 | |

Note: [a] Statistical significance $p<0.05$ versus control group. Data were expressed as mean ± SEM.
Примечание: [a] статистическая значимость $p<0,05$ по сравнению с контрольной группой. Данные выражали как среднее ± SEM

After 4−5 days of incubation at 37 °C in a $CO_2$-controlled humidified atmosphere, CEM giant (syncytium) cell formation was examined microscopically. The $EC_{50}$ (50% effective concentration) was defined as the compound concentration required to inhibit HIV-induced giant cell formation by 50%. Cytostatic activity assays were performed in 96-well microtiter plates [22]. To each well were added $(5\text{-}7.5)\cdot 10^4$ tumor cells and a given amount of the test compound. The cells were allowed to proliferate for 48 h (murine leukemia L1210 cells) or 72 h (human lymphocytic CEM and human cervix carcinoma HeLa cells) at 37 °C in a humidified $CO_2$-controlled atmosphere. At the end of the incubation period, the cells were counted in a Coulter counter. The $IC_{50}$ (50% inhibitory concentration) was defined as the concentration of the compound that inhibited cell proliferation by 50%.

## RESULTS AND DISCUSSION

A dose dependant effect of Compound **1** at 10 mg/kg on the Inhibitory avoidance test showed a significant antiamnesic action increasing latency by 167% and decreasing time spent in dark side by 56% and slight effect was found with 50 mg/kg.

The results shown in table 1 demonstrate that intraperitonealy administrated Compound **2** at both doses significantly decreased immobility and increased strong mobility (climbing behavior). The results suggest a significant antidepressant activity for Compound **2** as appears in the forced swim test. Additionally, Compound **2** demonstrated a significant latency increase and time spent in unsafe side decrease indicating antiamnesic action at both doses on the Inhibitory avoidance test (Table 2).

As table 1 indicates, Compound **3** induced a dose dependent increase in the horizontal and vertical motility of the animals. Compound **3** showed a marked increased exploratory behavior at 50 mg/kg. Data of the table 1 demonstrate that animals treated with Compound **3** (50 mg/kg) spent more time on the open arms and less on the center on the Elevated Pluz Maze test, suggesting a potential anxiolytic activity for the Compound. Compound **3** at the low dose (10 mg/kg) tended to increase the latency and decreased the time spent in dark side on the Inhibitory avoidance test (Table 2). These results suggest a potential antiamnesic activity for the Compound **3**.

Compound **4** at 50 mg/kg increased the horizontal behavior in the open field and decreased immobility in the Forced Swim Test (Table 1) as compared to vehicle treated animals. The results suggest a psycho stimulant action. On the other hand, Compound **4** at 10 and 50 mg/kg on the Inhibitory avoidance test demonstrated a significant latency increase (586% and 841%) and time spent in dark side decrease (70 and 100%, correspondingly) indicating good memory performance (Table 2).

From this study, it can be concluded that Compounds **3** and **4** marked analgesic activity (Table 2). In addition, our present investigation shows that all compounds are very safe for consumption with high $LD_{50}$ value (Table 2).

The present study also showed that synthesized compounds have neither antiviral activity nor cytotoxicity (minimum cytotoxic concentration > 100 μM) was observed.

In conclusion, we have synthesized novel water-soluble salts (Na+, Li+) of salicylamides which exhibited promising biological properties in combination with low toxicity. Disodium salicylurate showed a significant antiamnesic action. The results suggested a noticeable antidepressant activity and antiamnesic action for dilithium salicylurate and sodium salt of salicyloyl morphlide, a psychostimulant action for lithium salt of salicyloyl morphlide and dilithium gamma-(N-salicylamino)butyrate with pronounced antiamnesic property. As expected, lithium salts demonstrated higher psychotropic effects.

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*The authors declare the absence a conflict of interest warranting disclosure in this article.*

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