UNIVERSIDADE FEDERAL DO RIO GRANDE DO SUL

CURSO DE BIOMEDICINA

TRABALHO DE CONCLUSÃO DE CURSO EM BIOMEDICINA

INFLUÊNCIA DE TRAÇOS COMPORTAMENTAIS NA HIPERLOCOMOÇÃO INDUZIDA POR CAFEÍNA, ANFETAMINA, APOMORFINA E DIZOCILPINA EM CAMUNDONGOS.

CÍCERO RAFAEL LEÃO GARCIA

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ÍNDICE GERAL

RESUMO

Os transtornos de humor e o temperamento emocional (como medo e raiva) apresentam traços compartilhados, como proposto no modelo de Lara e colaboradores. Uma vez que medo e raiva são expressões que também podem ser observadas em animais, a utilização de situações comportamentais que possam analisar parâmetros relacionados com essas duas características pode ser útil para testar hipóteses acerca das suas bases biológicas bem como da sua relevância em transtornos do humor. O objetivo geral desse estudo foi investigar camundongos com baixa e alta atividade exploratória (LE e HE, respectivamente) em relação as suas diferenças neurobiológicas. Especificamente, o objetivo foi verificar se LE e HE diferem quanto à hiperlocomoção induzida por cafeína (30 mg/kg), anfetamina (1 e 2 mg/kg), apomorfina (2 mg/kg) e dizocilpina (0,25 mg/kg). Em relação ao período de habituação foram observadas diferenças significativas ($p < 0,05$; ANOVA de medidas repetidas) entre os grupos LE e HE, confirmando que camundongos LE e HE diferem quanto à procura por novidade. No entanto, não foram encontradas diferenças significativas entre os grupos LE e HE quanto à resposta à hiperlocomoção induzida por todas as drogas, nas doses utilizadas nesse estudo. Essa resposta equivalente indica que os sistemas em que essas drogas atuam (dopaminérgico, adenosinérgico e glumatérgico) não parecem contribuir de modo significativo para a diferença de fenótipo dos camundongos LE e HE, o que não exclui a possibilidade de que diferenças significantes possam ser caracterizadas ao se utilizar drogas que tenham ação em diferentes sistemas de neurotransmissores e/ou com a análise de comportamentos de maior complexidade e especificidade. Portanto, a continuidade deste estudo prevê o uso de outras drogas (opióides e serotonérgicas) e a análise de comportamentos mais especificamente associados ao medo e raiva, como a ansiedade, agressividade e dependência ao reforço.

Palavras-chave: temperamento, diferenças individuais, atividade no campo aberto, anfetamina, cafeína, dizocilpina, apomorfina

INTRODUÇÃO

1.1 Personalidade e comportamento

O conceito de personalidade vem sendo definido como "a organização dinâmica dos sistemas psicofísicos presentes no individuo que determinam, de forma única, sua adaptação ao ambiente" (Allport, 1937). Tendo em vista essa definição, modelos teóricos de personalidade geralmente são úteis na predição e tratamento de transtornos psiquiátricos, uma vez que visam a explicar o comportamento humano não só na sua normalidade, mas também nos quadros de transtornos psicológicos (Bond, 2001).

A busca por modelos que possam explicar de forma mais adequada o comportamento e os aspectos da personalidade humana não é um assunto recente na história da ciência. Muito tempo já se passou desde que Hipócrates, baseando-se em suas observações da fisiologia corporal, postulou suas idéias sobre medo e melancolia como traços do temperamento humano (Akiskal, 2005). Inúmeros modelos surgiram ao longo do tempo, dando suas contribuições para que algum dia se chegue a um entendimento maior sobre o comportamento humano como um todo e, a cada novo modelo que surge, as lacunas que antes eram deixadas pelos modelos anteriores vão eventualmente sendo preenchidas.

Lara e colaboradores (2006 a,b), tendo como base os modelos de Cloninger e Akiskal, propuseram um modelo teórico que classifica o espectro comportamental em um modelo bidimensional envolvendo dois eixos principais de modulação: medo e raiva (Figura 1).

Nesse modelo, o medo é responsável pela inibição do comportamento, agindo como um "freio" para as tomadas de decisões. Correlacionando com o modelo psicobiológico de temperamento (Cloninger, 1993), o medo está altamente relacionado à *evitação de dano* ou *Harm Avoidance* (HA), uma vez que ambos os traços apresentam uma predisposição à inibição ou término de comportamentos, o que está associado com pensamento pessimista, atitudes passivas e/ou preventivas como insegurança, medo do desconhecido, timidez, baixa energia e fadiga em excesso. Dessa maneira, indivíduos que apresentam baixo medo tendem a ser mais otimistas, confiantes, extrovertidos, enérgicos, seguros de si e, de uma maneira geral,

mais propensos a correrem riscos enquanto que, de maneira oposta, indivíduos com alto medo tendem a maior reclusão, timidez, preocupação e apresentam uma visão mais negativa do ambiente ao seu redor.

A raiva e a vontade/desejo seriam responsáveis pela ativação do comportamento e, novamente correlacionando com o modelo de Cloninger (1993), estão altamente relacionados à *procura por novidade* ou *Novelty Seeking* (NS). Ambos os traços envolvem impulsividade, irritabilidade, curiosidade e extravagância. Assim sendo, pessoas com alta raiva/vontade são mais impulsivas, dominantes, atentas, focadas e passionais, ao contrário de pessoas com baixa raiva/vontade que apresentam características mais passivas, tendendo a um comportamento mais calmo, beirando à apatia em determinados casos.

Uma vez que medo e raiva se apresentam como dois eixos dimensionais independentes, qualquer combinação entre eles é possível e, desse modo, se obtêm um amplo espectro do comportamento humano (Figura 1).
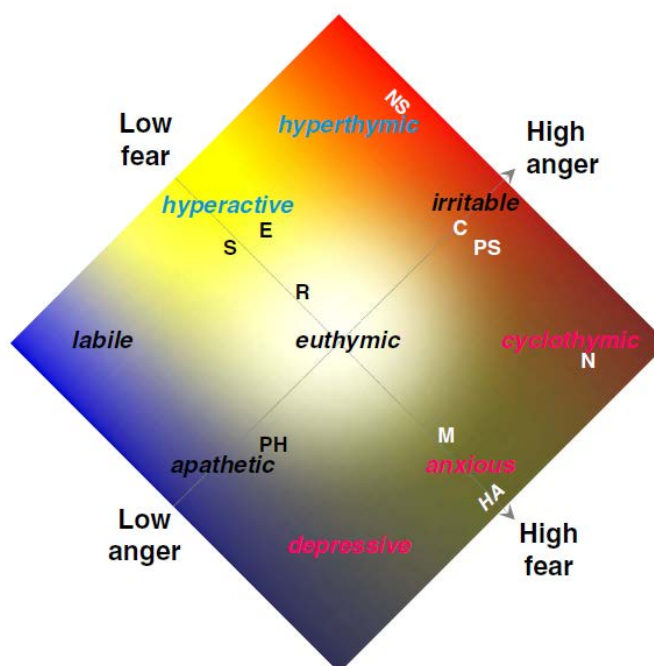

Figura 1: Modelo bidimensional de medo e raiva e temperamentos afetivos. Retirado de *Lara DR et al., 2006 – Toward an integrative model of the spectrum of mood, behavioral and personality disorders based on fear and anger traits: I. Clinical implications*

Um temperamento mais irritadiço, por exemplo, pode ser entendido como resultado de alta raiva combinada com um traço moderado de medo; já um

temperamento mais ansioso poderia resultar da combinação de traços de personalidades de alto medo com raiva moderada.

1.2.1 Bases neurobiológicas do medo e da raiva

Medo e raiva são emoções bem conservadas evolutivamente entre as espécies (Cloninger, 1993) e estão envolvidas com as áreas límbicas, que servem principalmente como forma de modulação das atividades mentais tanto superiores como inferiores. A amígdala é um núcleo subcortical responsável entre outras funções pelo reconhecimento do medo (Garret e Chang, 2008), possuindo extensa comunicação com áreas sensoriais do cérebro, incluindo aferências visuais, auditivas, somato-sensorias e gustatórias, e eferências para áreas responsáveis por respostas autonômicas como aumento das funções cardíaca e respiratória. Em termos de raiva/vontade, outras áreas particularmente relevantes incluem a área tegmentar ventral, o núcleo accumbens e o córtex frontal.

Em relação à participação de neurotransmissores, o medo é altamente influenciado pelos sistemas serotoninérgico, noradrenérgico e GABAérgico, enquanto que a raiva/vontade parece ser predominantemente regulada pelos sistemas dopaminérgico e glutamatérgico (Figura 2) (Lara et al., 2006 b).
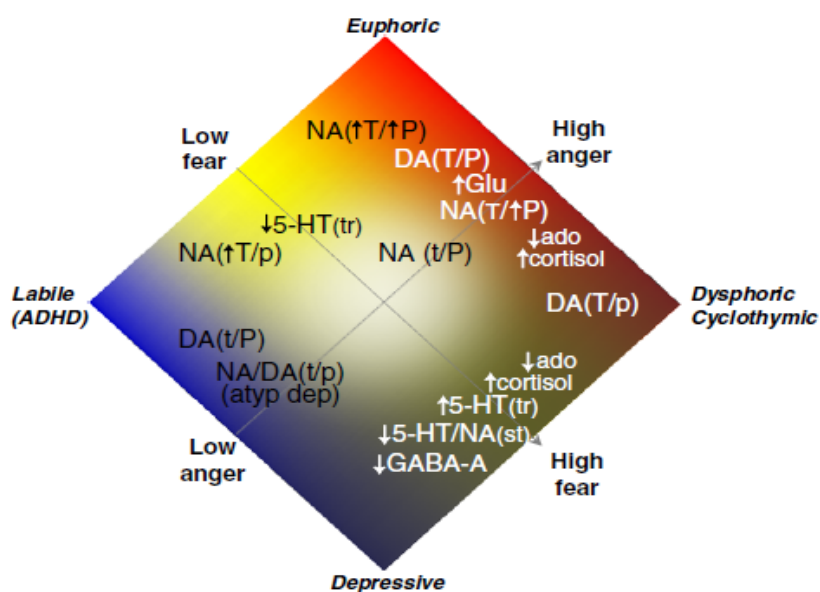

Figura 2: Correlações neuroquímicas de medo e raiva. Retirado de *Lara DR et al., 2006 b – Toward an integrative model of the spectrum of mood, behavioral and personality disorders based on fear and anger traits: II Implications for neurobiology, genetics and psychopharmacological treatment*

Outros mediadores químicos também parecem ter relação com medo e raiva; por exemplo, a adenosina possui atividade no sistema nervoso central e pode induzir uma gama de sintomas tais como ansiedade e hiperatividade (Bond 2001).

1.2.2   Medo e raiva em transtornos psicológicos

De acordo com o modelo de Lara e colaboradores (2006 a,b), traços de medo e raiva/vontade podem se encontrar desregulados ou acentuados nos indivíduos acometidos por transtornos de personalidade e de humor. Da mesma forma, alguns distúrbios psicológicos seriam influenciados pelo padrão de comportamento normal do indivíduo.

Em quadros de mania, por exemplo, alguns sintomas como comportamento de alta energia, temperamento expansivo, alta impulsividade e tendência a ações de risco podem ser interpretados principalmente como um traço intenso de baixo medo, enquanto que a tendência à irritabilidade, procura por satisfação e objetividade de idéias estaria mais relacionado à alta raiva. Episódios de depressão, tristeza, anedonia, fadiga e sentimentos de desvalia seriam derivados de alto medo, enquanto que a apatia e o retardo psicomotor estariam associados à baixa raiva.

Analisando os diferentes transtornos de personalidade e humor e fazendo a transposição para o modelo de medo e raiva, foi possível propor que muitos desses transtornos apresentam traços compartilhados (Figura 3): transtorno de personalidade borderline, cleptomania e transtorno de personalidade anti-social são exemplos de distúrbios que apresentam em comum altos índices de raiva. Esse traço comum poderia explicar por que medicamentos indicados para tratar um tipo específico de transtorno também apresentam resultados positivos ao serem utilizados em outros tipos de transtorno.
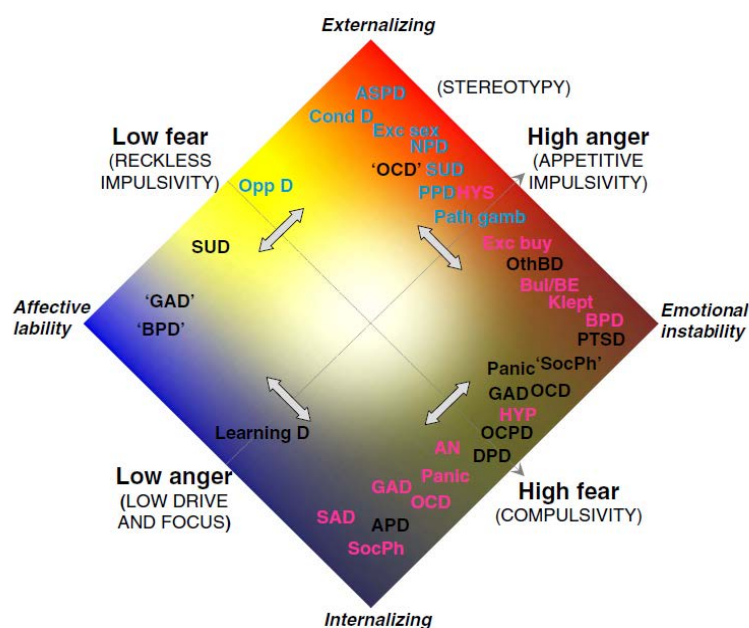
Figura 3: Medo e raiva em transtornos comportamentais e de personalidade ASPD = antisocial personality disorder, Opp. D = oppositional defiant disorder; Exc sex = excessive/eccentric sexual behavior, Conduct D = conduct disorders, PPD = paranoid personality disorder, SUD = substance (ab)use disorder, Path gamb = pathological gambling, Exc buying = excessive buying or shopping, NPD—narcissistic personality disorder, Hys = histrionic personality disorder, OthBD = other behavioral disorders (e.g. excessive ⬜mygdale use, self-inflicting behavior, skin picking, pyromania, pedophilia, antisocial acts, intermittent explosive disorder, excessive skin tanning, Tourette syndrome, autism), Bul/BE = bulimia and binge eating, Klept = kleptomania, BPD = borderline personality disorder, PTSD = post-traumatic stress disorder, SocPh = social phobia, OCD = obsessive compulsive disorder, Panic = panic attacks and disorder, GAD = generalized anxiety disorder, BDD—body dysmorphic disorder, HYP = hypochondriasis, AN = anorexia nervosa, OPD = obsessive personality disorder, DPD = dependent personality disorder, APD — avoidant personality disorder, SAD — separation anxiety disorder. 'BPD', 'GAD', 'OCD' and 'SocPh' denote atypical forms of the disorder. Note that the traditional "compulsive" nomenclature was substituted for "excessive" if related to high anger (e.g. excessive buying or sexual behavior). . Retirado de *Lara DR et al., 2006 a – Toward an integrative model of the spectrum of mood, behavioral and personality disorders based on fear and anger traits: I. Clinical implications*

1.2.3. Medo e raiva em animais

Uma vez que medo e raiva/vontade não são características exclusivamente humanas, a observação desses comportamentos em animais e a utilização de testes que possam analisar parâmetros de comportamento relacionados com essas características comportamentais são extremamente úteis para testar hipóteses acerca das bases biológicas do medo e da raiva, sua relevância em transtornos psicológicos e o efeito de psicofármacos.

De acordo com Kazlauckas e colaboradores (2005) a atividade exploratória no teste de campo aberto com objeto central serve como modelo para diferenciar

camundongos de acordo com padrões de comportamento que representam diferentes temperamentos: o estudo correlacionou o desempenho de animais no campo aberto com níveis de ansiedade e agressividade verificados em modelos comportamentais relevantes. De acordo com os resultados obtidos, animais que apresentam uma maior taxa de exploração ao objeto no campo aberto apresentaram maior procura por novidade, baixa evitação ao dano e alta agressividade, enquanto que os animais com menor taxa de exploração ao objeto no campo aberto apresentaram comportamento exatamente oposto, com menor procura por novidade, alta evitação ao dano, baixa agressividade e em alguns casos comportamento submisso. Desse modo, animais com alta atividade exploratória possuem características que podem defini-los como tendo baixo medo e alta raiva/vontade, enquanto que os animais com baixa atividade exploratória apresentam alto medo e baixa raiva/vontade.

1.3.   Modelos animais de mania e psicose

O uso de drogas psicoativas em modelos animais é uma maneira de se avaliar as bases neuroquímicas de diversos tipos de doenças. Por exemplo, a utilização de anfetamina, que entre outros efeitos induz hiperatividade motora ao promover a liberação de dopamina, é um dos modelos animais relacionados à mania. Outros modelos experimentais de mania comumente utilizados incluem a administração aguda de cafeína, apomorfina e dizocilpina: cafeína atua como um antagonista não seletivo de receptores adenosinérgicos do tipo A_1 e A_{2A}; apomorfina é um agonista dopaminérgico não seletivo que se liga a receptores D_1 e D_2; dizocilpina é um antagonista de receptores glutamatérgicos do tipo NMDA (n-metil-d-aspartato). (REFS)

2. OBJETIVOS

2.1 Objetivos gerais

Considerando alta raiva/vontade e baixo medo como características predominantes em pacientes que apresentam quadros de mania, e que baixa raiva/vontade e alto medo são mais freqüentemente observados em pacientes com depressão, aliado ao grande interesse na modelagem animal desses transtornos, o objetivo geral desse estudo foi dar início à identificação de eventuais diferenças neurofarmacológicas em camundongos com alta e baixa atividade exploratória.

2.2 Objetivos específicos

Verificar se camundongos com baixa e alta atividade exploratória diferem quanto à hiperlocomoção induzida por anfetamina, cafeína, apomorfina, e dizocilpina.

3. ARTIGO CIENTÍFICO

Leão Garcia, CR; Kazlauckas, V; Elisabetsky, E; Lara, DR; **Do mice with distinct behavioral patterns respond differently to psychoactive drugs? I. Response to caffeine-, amphetamine-, apomorphine- and dizocilpine-induced hyperlocomotion.**

Do mice with distinct behavioral patterns respond differently to psychoactive drugs? I. Response to caffeine-, amphetamine-, apomorphine- and dizocilpine-induced hyperlocomotion.

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Abstract

Recently, Lara et al. (2006 a,b) proposed a theoretical model in which behavior can be organized in a bidimensional model involving two axis: fear and anger/drive, in which fear is responsible for inhibitory behaviors, acting as a "brake" in decision making, whereas anger/drive initiate behavior. Fear is highly correlated with harm avoidance (HA), since pessimism, fear of the uncertain, timidity and low energy or fatigability are characteristics related with both fear and HA. Because fear and anger can also be observed in animals, the analysis of these two behavioral traits can be useful for testing hypothesis on the biological basis for fear and anger, and its relation to human mood disorders. The purpose of this study was to verify if mice with low and high exploratory activity (LE and HE) differ in regard to the hyperlocomotion induced by amphetamine, caffeine, dizocilpine, and apomorphine. Significant differences between LE and HE groups regarding habituation were seen in 4 out of 6 experiments, confirming that LE and HE mice differ in novelty-seeking behavior. However no significant differences were observed in the response of LE and HE mice in regard to the hyperlocomotion induced by any of the drugs and dosages here used. This equivalent responses indicate that the sensitivity of these systems in LE and HE mice is not clearly distinct. Further studies using drugs acting on other neurotransmitter systems, or other behavioral analyses of greater complexity and better specificity than locomotor activity should be performed to better study the neurobiological differences LE and HE mice.

Keywords: temperament, individual differences, open-field activity, anfetamine, caffeine, dizocilpine, apomorphine

1. Introduction

The search for psychological models that could more adequately explain human behavior and aspects of human personality has been present throughout the science history. Recently, Lara et al. (2006 a,b) proposed a theoretical model in which the behavioral spectrum is organized in a bidimensional model involving two axis: fear and anger/drive. In this model, fear is responsible for inhibitory behaviors, acting as a "brake" in decision making. Fear is highly correlated with harm avoidance (HA), since pessimism, fear of the uncertain, timidity and low energy or fatigability are characteristics related with both fear and HA. Individuals presenting low fear are usually more optimistic, confident, outgoing and energetic, more likely to undertake risks, while individuals with high fear tend to be more recluse, shy, concerned and usually present a more negative life perspective. Anger/drive would be responsible for initiating behaviors, being highly correlated with novelty seeking (NS). Both anger and NS involve impulsivity, irritability, curiosity, extravagance and search for immediate gratification. Individuals with high anger tend to be more impulsive, authoritative, focused and passionate, unlike those characterized by low anger/drive, who tend to be more passive, calm and, in some cases, apathetic. Since fear and anger/drive are in this model two interdependent axis, many combinations are possible, representing a wide spectrum of human behaviors as well as related psychopathologies.

Mood disorders have shared traits with temperaments characterized in terms of fear and anger/drive. For instance, some of the symptoms of mania, like expansive mood, high energy, pressure to talk and risk taking behavior can be interpreted as of very low fear, whereas irritable, pleasure-seeking and goal directed behavior would be better associated with excessive anger. In depressive episodes, the commonly observed symptoms of sadness, anhedonia and worthlessness could be correlated with high fear, while apathy and psychomotor retardation are associated with very low anger.

Evolutionarily, fear and anger are well conserved emotions (Cloninger, 1993), related to the limbic system. The amygdale is the subcortical area responsible for fear recognition (Garret & Chang, 2008), effectively communicating with sensory areas of the brain (including afferents from visual, auditory, somatosensory, and

gustatory afferents) and the autonomic nervous system (efferent to brain areas modulating heart and respiration rates). Regarding anger, particularly relevant areas are the ventral tegmental area, the nucleus accumbens and the frontal cortex.

Regarding the neurochemical basis of these two emotional dimensions, fear is much influenced by the serotonergic, noradrenergic and GABAergic systems, whereas anger seems to be mostly regulated by the dopaminergic (associated with rewarding and novelty seeking) and glutamatergic systems (Lara et al., 2006b). Other modulatory neurotransmitters are likely to be related to fear and anger, including adenosine (which induces anxiety and hyperactivity).

Since fear and anger/drive can be observed in animals, the utilization of tasks to analyze parameters related with these two traits can be useful for testing hypothesis on biological basis for fear and anger, and its relation to human mood disorders. According to Kazlauckas et al. (2005), the exploratory activity in an open-field with a central object is an adequate strategy to differentiate mice according to behavioral patterns that represent temperament types: animals with a higher object exploration rate – represented as the time spent in the central area of the open-field – showed more behavioral traits of NS, less HA and higher aggressiveness, whereas animals with low exploratory activity rates show the opposite behavior: less NS, high HA and low aggressiveness. Therefore, mice with high exploratory activity can be classified as having low fear and high anger/drive, while mice showing low exploratory behavior can be classified as having high fear and low anger/drive.

Considering the trait similarities in terms of fear and anger/drive of mood disorders such as mania and depression, as well as the usefulness of modeling these disorders in animals, characterizing neuropharmacological differences in mice presenting low and high exploratory behavior can be of interest. Since noradrenaline, dopamine and adenosine have been associated with fear, anger, HA and NS, the purpose of this study was to verify if mice with low and high exploratory activity differ in regard to the hyperlocomotion induced by amphetamine, caffeine, dizocilpine, and apomorphine.

2. Methods

2.1 Animals

Experiments were performed with 80 CF1 male adult mice (8 weeks old, 35-45 g), housed six to eight per cage with food and water *ad libitum*, under controlled laboratory conditions (12 h light/dark cycle, lights on at 8:00 am, $21 \pm 2 \ ^{o}C$). All animal experiments were conducted between 9:00 am and 6:00 pm. Experiments and animal handling were in accordance with institutional animal care guidelines and the principles of laboratory animal care as stated in the Guide for the Care and Use of Laboratory Animals. Efforts were made in order to use the minimal amount of mice necessary for experiments and reduce their suffering. Before every experiment the mice were habituated to the experimental room for 30 min prior to the experiment.

2.2 Behavioral separation of high and low exploratory mice

2.2.1 Open-field with a central object

This test was used to separate two extreme mice populations regarding exploration of an object in a new environment (Kazlauckas, 2005). The animal was placed in an open-field (50 cm × 50 cm × 50 cm) with an object (a white cylinder of 1.5 cm radius and 6 cm high) placed on the center of the arena to stimulate exploration. Exploratory behavior (the time spent by the animal in and out of an imaginary center square of 30 cm × 30 cm) was recorded for 5 min and analyzed by ANY-MAZE software. Eighty mice were screened, and 24 animals were selected from each extreme of exploratory behavior (the 12 least and the 12 most explorers) to compose the low (LE) and high (HE) exploratory groups, respectively. Mice remained in their home cages without changing housemates until the end of all the experiments.

2.3 Pharmacological treatment and locomotor activity

2.3.1 Open-field

Mice were placed in the same open-field (except that object free) apparatus as above and the locomotor activities of four mice were analyzed simultaneously, being two LE and two HE mice. The distance traveled by each animal was calculated by ANY-MAZE for 150 min. The first 60 min were considered as habituation; following this period mice received an i.p. injection (10 ml/kg) of saline (0.9% NaCl, LE/HE n=12), amphetamine (1.0 mg/kg, LE/HE n=12 ; 2.0 mg/kg, LE/HE n=12), dizocilpine (0.25 mg/kg, LE/HE n=10), caffeine (30 mg/kg, LE/HE n=10) or apomorphine (2 mg/kg, LE/HE n=10) and were immediately back to the apparatus for another 90 min, considered as locomotion. At least one drug-free week was allowed between experiments. All drugs used were purchased from Sigma/RBI (St. Luis, MO, USA).

2.4 Statistical Analyses

Prisma Graph-Pad was used for the statistical analyses. Student's t-test was used to analyze the percentage of time spent in the center of the open-field of LE and HE mice. To evaluate the effects of drugs in habituation and locomotion repeated measures analysis of variance (ANOVA-RM) was used, with time as the repeated measure and temperament as the independent variable. Sudent's t-test was used to compare LE and HE locomotor activity at a given time. $p<0.05$ was adopted for significance.

3. Results

The mean percentage of time spent in the center area of the open field (mean ± S.D.) for the whole group (n=80) was 37.9±8.9%; the higher and lower cut-off values for the top and low activity groups of 12 mice were >47% and <29.3%. The mean percentage of time spent in the center of the open field was significantly different (t=13.37, p<0.0001) for LE and HE, with 23.9±6% for LE and 49.1±2.6%

for HE (Fig. 1A). There was no difference in the locomotor activity (total distance) between LE and HE (Fig. 1B) (t=1.203, p>0,05). The remaining 56 animals (those having between 29.3% and 47% of the time spent in the center of the open-field) were kept throughout the experiment in order to maintain the home cages for all animals.

The effect of treatments on locomotor activity for LE and HE is shown in Fig. 2. Regarding the habituation (pre-drug) period, ANOVA-RM showed significant differences between LE and HE in pre-saline ($F_{1,110}=4.47$; $p<0.05$), pre-amphetamine 1 mg/kg ($F_{1,110}=4.42$; $p<0.05$), pre-amphetamine 2 mg/kg ($F_{1,110}=5.77$; $p<0.05$), pre-apomorphine ($F_{1,90}=5.46$; $p<0.05$),but not in pre-dizocilpine ($F_{1,90}=1.76$; $p>0.05$) and pre-caffeine ($F_{1,90}=3.59$; $p>0.05$). T-test showed that such differences were due to limited time points. Regarding the locomotion period, no significant differences were found between LE and HE following treatment [saline ($F_{1,176}=1.42$; $p>0.05$), amphetamine 1 m/kg ($F_{1,176}=0.02$; $p>0.05$), amphetamine 2 mg/kg ($F_{1,176}=1.52$; $p>0.05$), dizocilpine ($F_{1,144}=0.35$; $p>0.05$), caffeine ($F_{1,144}=1.39$; $p>0.05$), apomorphine ($F_{1,144}=0.21$; $p>0.05$)].

4. Discussion

Significant differences between LE and HE groups regarding habituation were seen in 4 out of 6 experiments. Since in this initial period mice were adapting to the environment, it is not surprising that the particular characteristics in exploratory activity that characterize LE and HE mice would lead to different habituation rates. These results are in agreement with Kazlauckas et al. (2005), confirming that LE and HE mice differ in novelty-seeking behavior. In studies with rats, in which animals are classified according to their response to a new environment (low response and high response, LR and HR respectively) this same difference in habituation was observed (Piazza et al., 1989, 1991; Gingras & Cools, 1997; Antoniou et al., 2004). As observed by Kazlauckas et al. (2005) and confirmed in this study, after habituated to a new environment LE and HE mice are no longer different in regard to locomotion.

No significant differences were observed in the response of LE and HE mice in regard to the hyperlocomotion induced by any of the drugs and dosages here used. This equivalent response may indicate that the sensitivity of the systems modulated by these drugs is not clearly distinct in LE and HE mice

In relation to amphetamine, Piazza et al. (1989, 1991) reported that, when exposed to doses of 0.3 and 1.5 mg/kg, HR rats showed a greater response in locomotor activity compared to LR rats. Gingras & Colls (1997), in a dose response study with amphetamine, also found a significant difference with the dose 1.5 mg/kg, but no significant differences with 0.5, 1.0 and 2.0 mg/kg. Nevertheless, Antoniou et al. (2004) using the same amphetamine dose of 1.5 mg/kg did not find differences in locomotor activity between LR and HR rats in response to amphetamine. Although differences in methods for differentiating LR/HR and LE/HE exist, as well as potential differences in rats and mice, taken together this data suggest that exploratory behavioral traits are not well correlated with the sensitivity of these animals to amphetamine.

Differences in the behavioral substrates for diverse psychopathologies as well as differences in the outcome of clinical responses to medication could suggest different baselines of specific neuronal circuitry and/or different influences of neurotransmitters systems. Regarding the anger dimension, Post (1995) showed that dopamine D_2 antagonists reduce aggressiveness, novelty seeking and mania symptoms. It is also reported that medications acting on glutamatergic pathways, such as the AMPA receptor blocker topiramate, can be useful in treating some impulsive or anger-related behaviors. In relation to fear, Joyce et al. (1994) examined the Cloninger's model and its ability to predict the outcome of patients with major depressive disorder. It was found that patients with high harm avoidance and reward dependency had better chance of a favorable response to treatment with either clomipramine or desipramine. There are also drugs that seem to modulate both fear and anger: according to Fredholm et al. (2005), caffeine can induce or aggravate a range of symptoms from both the fear and anger domains, such as anxiety, panic attacks and possibly hypomania or mixed states.

Despite the results found in this initial approach, we can not reject the possibility of different results by using other types of neurotransmitter systems. Accordingly, Antoniou et al. (2008) through neurochemical analyzes reported

significant differences in serotonin levels at cortical areas of LR compared to HR, with HR showing a reduced serotoninergic turnover ratio in the prefrontal cortex than LR. The use of behaviors of greater complexity and better specificity than locomotor activity may also reveal differences between LE and HE. For instance, it has been documented that HR rats have a higher propensity to self administration for amphetamine (Piazza et al. 1989, 1991; Kleuber et al. 2001) and cocaine (Davis et al. 2008).

5. Conclusion

In this first approach of our study no obvious differences were found, which does not exclude the possibility that different results would have been obtained with drugs acting on other neurotransmitter systems or different doses of the same drugs. It is important to consider that the use of behaviors of greater complexity and better specificity than locomotor activity may reveal differences between LE and HE. Therefore, analyses of behaviors more specifically associated with fear and anger/drive, such as anxiety, aggressiveness, and response to rewards such as palatable food and/or sucrose consumption, are warranted.

6. Acknowledgments

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Figure 1: Performance of low and high exploratory mice in the open-field with a central object. (A) Percentage of time spent in central area. Dots represent individual mice (white, low exploratory; gray, high exploratory) and dash represents mean values. **p<0.001 (Student's t-test). (B) Total distance traveled during the experiment. Data expressed as mean±S.D.

Figure 2: Locomotor activity of high and low exploratory mice acutely treated with (A) saline, (B) amphetamine (1 mg/kg), (C) amphetamine (2 mg/kg), (D) dizocilpine (0.25 mg/kg), (E) caffeine (30 mg/kg) or (F) apomorphine (2 mg/kg) after 60 min of habituation period in a novel environment. Data expressed as mean± S.E.M. *=p<0.05 (Student's t-test).

Figure 1

A - Saline

B – Amphetamine 1 mg/kg

C – Amphetamine 2 mg/kg

D – Dizolcipine 0.25 mg/kg

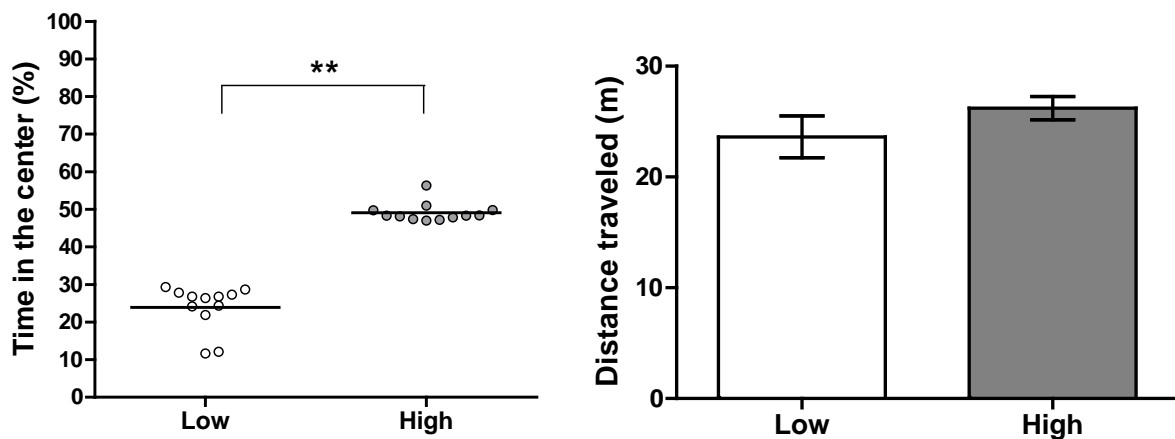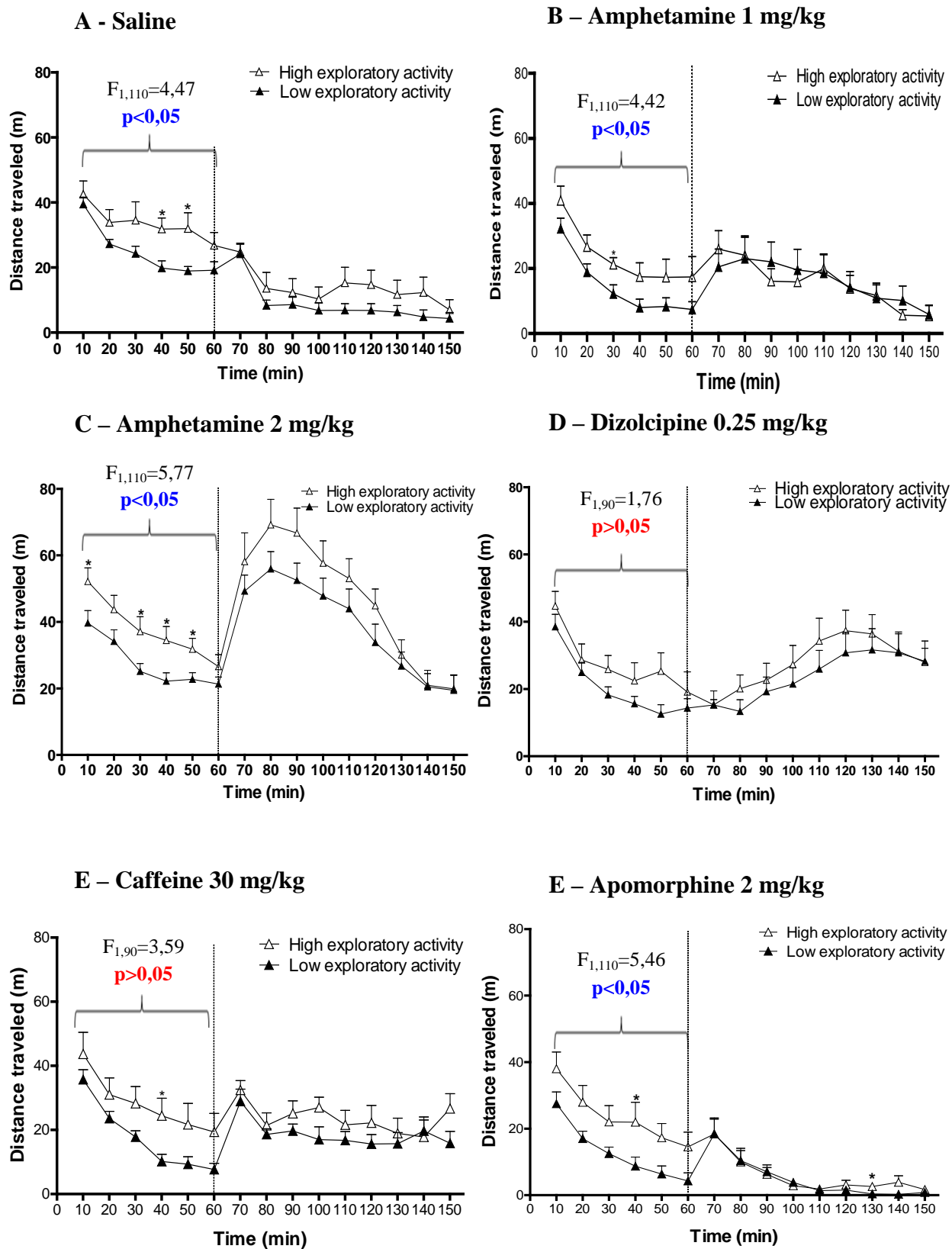**E – Caffeine 30 mg/kg**

E – Apomorphine 2 mg/kg

Figure 2

4. CONCLUSÃO E PERSPECTIVAS

Iniciamos com esse trabalho a investigação da relação comportamental de roedores na resposta a modelos animais de psicopatologias. A triagem inicial se mostrou eficaz em separar os animais de acordo com suas características comportamentais. Foi possível separar camundongos em dois grupos que apresentaram características distintas na atividade exploratória. No entanto, a administração de drogas comumente utilizadas em modelos de hiperlocomoção não mostrou qualquer diferença entre os grupos quanto ao aumento da atividade locomotora induzida em camundongos separados por baixa e alta atividade exploratória.

Os resultados encontrados não excluem a possibilidade de que diferenças significativas entre os dois grupos de camundongos possam ser obtidas ao se utilizar drogas que tenham ação em diferentes sistemas de neurotransmissores ou ao se utilizar comportamentos de maior complexidade e melhor especificidade. Portanto, análise de comportamentos mais especificamente associada ao medo e raiva, como a ansiedade, agressividade e dependência ao reforço, são necessários.

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6. ANEXO - Normas do Periódico Progress in Neuro-Psychopharmacology & Biological Psychiatry

Guide for Authors

Specific recommendations for original articles

1. Title page. This should contain:

1.1. Complete title of the article. The title of the paper should be brief; no longer than 100 characters in length, and should capture and communicate the key message of your research to a broader audience. To aid this, abbreviations, unless familiar to a broad audience, should be avoided

1.2. Names of all authors, with an asterisk beside the name of the corresponding author. Where the family name may be ambiguous (e.g. a double name), please indicate this clearly.

1.3. Full mailing addresses of each author, including the name of the institution and the department. Present the Authors' affiliation addresses (where the actual work was done) below the names. If an Author has moved since the work described in the article was done, or was visiting at the time, a "Present address" (or Permanent address") may be indicated as a footnote to that Author's name.

1.4. Links (lowercase roman letters) connecting authors with their affiliations (all of the authors' names should be on one line and all of their affiliations on another; if authors share an affiliation, they should share the link).

2. Abstract page. This should contain:

2.1. Abstract representing in concise form the purpose, the general methods, the findings and the conclusions of the authors.

2.2. Keywords in alphabetical order, given as an aid to indexing.

2.3. Abbreviations. Whenever an abbreviation other than those listed under General recommendations is used in an article, it is to be defined in text at first mention. An alphabetical list of abbreviations, followed by their full terms, should be placed under the keywords

3. Body of the text

3.1. Introduction. This section should contain a clear statement of the general and specific objectives as well as the hypotheses which the work is designed to test. It should also give a brief account of the reported literature. The last sentence should clearly state the purpose of the article.

3.2. Methods. This section should contain explicit, concise descriptions of all procedures, materials and methods used in the investigation to enable the reader to judge their accuracy, reproducibility, etc. To increase clarity, headings should be used throughout. For example, the following subheadings, which should be numbered, could be used:

3.2.1. Experimental articles (full length or short papers): Animals, Drugs, Apparatus, Experimental procedure, and Statistical analysis.

3.2.2. Clinical articles (full length or short papers): Patient population, Drug administration, Study design, Assessment instruments, and Data analysis. Depending on the type of article they are preparing, authors could introduce any other subheadings they find useful.

3.3. Results. This section usually contains the experimental data, but no extended discussion of their significance. The results should be illustrated (figures and tables); data are usually easier for readers to grasp if they are represented in graphic or tabular form, rather than discursively. Graphic presentation of data is preferred. Data should not be needlessly repeated in text. Sufficient data may allow interested but non-expert readers to judge the variability and reliability of the results. The section should be well structured using appropriate subheadings.

3.4. Discussion. This should be pertinent to the results. Speculative discussion is not discouraged provided it is based on the data presented. The discussion should be as concise as possible and well structured, using appropriate subheadings.

3.5. Conclusions. A short paragraph of conclusions (5 to 10 lines) should be included.

4. Acknowledgments. These may be included at the end of the text before the References; they should have a separate heading.

5. References.

5.1 *Text:* All citations in the text should refer to:

1. *Single author:* the author's name (without initials, unless there is ambiguity) and the year of publication;

2. *Two authors:* both authors' names and the year of publication;

3. *Three or more authors:* first author's name followed by 'et al.' and the year of publication. Citations may be made directly (or parenthetically). Groups of references should be listed first alphabetically, then chronologically.

Examples: "as demonstrated in wheat (Allan, 1996a, 1996b, 1999; Allan and Jones, 1995). Kramer et al. (2000) have recently shown...."

5.2 *List:* References should be arranged first alphabetically and then further sorted chronologically if necessary. More than one reference from the same author(s) in the same year must be identified by the letters "a", "b", "c", etc., placed after the year of publication.

Examples:

5.2.1 Reference to a journal publication:

Van der Geer J, Hanraads JAJ, Lupton RA. The art of writing a scientific article. J Sci Commun 2000;163:51-9.

5.2.2 Reference to a book:

Strunk Jr W, White EB. The elements of style. 3rd ed. New York: Macmillan, 1979.

5.2.3 Reference to a chapter in an edited book:

Mettam GR, Adams LB. How to prepare an electronic version of your article. In: Jones BS, Smith RZ, editors. Introduction to the electronic age. New York: E-

Publishing Inc.; 1994.p.281-304.

Note shortened form for last page number. e.g., 51-9, and that for more than 6 authors the first 6 should be listed followed by 'et al.' For further details you are referred to "Uniform Requirements for Manuscripts submitted to Biomedical Journals" (J Am Med Assoc 1997;277:927-34), see also ➭ http://www.nlm.nih.gov/tsd/serials/terms_cond.html

6. Tables. All tables must be cited in the text, have brief, descriptive titles and be consecutively numbered with Arabic numerals. Information other than that defining the data should be presented as footnotes. Use lowercase roman letters for footnotes. Only horizontal rules should be included, and kept to a minimum.

7. Illustrations. Each illustration should be clearly marked on the reverse side with the name of the corresponding author, the number of the illustration and its orientation (top); use a soft pencil or felt-ripped pen, and do not press hard against the surface.

7.1. Photographs. Photographs should be glossy prints with high contrast. Magnification should be indicated by a line representing the actual scale of reproduction (0.1 μm, 1 μm). Avoid the use of magnification factors whenever possible.

7.2. Line figures. Figures will not be redrawn by the Publisher. They should be black ink on white paper, or black and white prints. Do not place the title of the figure within the figure itself. The size of the lettering should be consistent, taking into consideration the possibility of reduction.

7.3. Colour figures. If together with your accepted article, you submit usable colour figures, then Elsevier will ensure, at no additional charge, that these figures will appear in colour on the Web (e.g., ScienceDirect and other sites) regardless of whether these illustrations are reproduced in colour in the printed version. For colour reproduction in print, you will receive information regarding the costs from Elsevier after receipt of your accepted article. For further information on the preparation of electronic artwork, please see ➭ http://www.elsevier.com/artworkinstructions [Please note: Because of technical complications that can arise in converting colour figures

to "grey scale" (for the printed version should you not opt for colour in print), please submit in addition usable black-and-white files corresponding to all the colour illustrations]. Authors should note that a request to revert from full colour to colour only in the electronic publication at the stage of typesetting and proof correction, will require separate editorial agreement, with possible re-review if necessary, and may significantly delay publication of your manuscript.

8. Legends for figures. These should be typed on a separate page, double spaced as part of the text. Legends, should be numbered consecutively in Arabic numerals. Legends should explain the figures in sufficient detail so that repeated referral to the text is unnecessary. Abbreviations in the legends should conform to those in the text.

9. Footnotes. These are best avoided or they should be kept to a minimum. When used, they should be typed at the bottom of the appropriate page and separated from the text by a short line. Footnotes should be used for authors' degrees and positions, proprietary names and trademarked drugs and other materials not appropriately referred to in the text or in the reference list.

10. Supplementary material: Electronic supplementary material is now accepted to support and enhance your scientific research. Supplementary files offer the author additional possibilities to publish supporting applications, movies, animation sequences, high-resolution images, background datasets, sound clips and more. Supplementary files supplied will be published online alongside the electronic version of your article in Elsevier web products, including ScienceDirect: ⊞⃗ http://www.sciencedirect.com. In order to ensure that your submitted material is directly usable, please ensure that data is provided in one of our recommended file formats. Authors should submit the material in electronic format together with the article and supply a concise and descriptive caption for each file. For more detailed instructions please visit our Corporate Website at ⊞⃗ http://www.elsevier.com/authors.

General recommendations

Abbreviations

Define abbreviations that are not standard in this field at their first occurrence in the article: in the abstract but also in the main text after it. Ensure consistency of abbreviations throughout the article.

The following abbreviations or their properly prefixed multiples and submultiples may be used without definition in the text, tables or figures (Notice to contributors, 1981, *J. Pharmacol. Exp. Ther.*):

Drug nomenclature

Generic names should be used in text, tables and figures. Trade names and the name and city of their manufacturer may be mentioned in parentheses in the first text reference to the drug, but should not appear in titles, figures or tables. Chemical names could also be used. Code numbers could be given in brackets. When a trade name is used, it should be capitalized; general or chemical names are not capitalized. The chemical nature of new drugs must be given when known. The form of drug used in calculations of doses (e.g., base or salt) should be indicated.

Studies on natural products The journal does *not* publish work on the actions of biological extracts unless the pharmacological active molecular substrate and/or specific receptor binding properties of the extract compounds are elucidated.

Ethical Standards

• The authors declare that all experiments on human subjects were conducted in accordance with the Declaration of Helsinki ⊞►http://www.wma.net and that all procedures were carried out with the adequate understanding and written consent of the subjects.

• The authors also certify that formal approval to conduct the experiments described has been obtained from the human subjects review board of their institution and could be provided upon request.

• If the studies deal with animal experiments, the authors certify that they were carried out in accordance with the National Institute of Health Guide for the Care and Use of Laboratory Animals (NIH Publications No. 80-23) revised 1996 or the UK Animals (Scientific Procedures) Act 1986 and associated guidelines, or the European Communities Council Directive of 24 November 1986 (86/609/EEC).

• The authors also certify that formal approval to conduct the experiments described has been obtained from the animal subjects review board of their institution and could be provided upon request.

• The authors further attest that all efforts were made to minimize the number of animals used and their suffering.

• If the ethical standard governing the reported research is different from those guidelines indicated above, the authors must provide information in the submission cover letter about which guidelines and oversight procedures were followed.

• The Editor reserves the right to return manuscripts in which there is any question as to the appropriate and ethical use of human or animal subjects.