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·调查·

骨肿瘤动物实验报告现状研究

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摘 要 目的 评价有关骨肿瘤动物实验研究的报告质量。**方法** 计算机检索中国知网、中国生物医学文献数据库、维普及万方数据 4 大中文数据库,检索时限均从 2010 年 1 月至 2012 年 12 月,检索文献语言为中文,纳入关于骨肿瘤动物实验的研究文献,再根据 ARRIVE 指南评估每一篇文献,最后用 SPSS 软件对评估所得的数据进行统计学分析。**结果** 最终纳入符合标准的骨肿瘤动物实验研究文献有 71 篇,每一篇文献的平均得分为(21.08±2.12)分,纳入文献报告质量普遍中等偏低。**结论** 我国骨肿瘤的动物实验报告质量总体来说很不理想,存在严重的潜在偏倚现象,未来的研究中应重视规范动物实验的设计及报告,以确保研究质量。

关键词 骨肿瘤;动物研究;报告
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The Study of the Report about Bone Tumor in Animal Experiment

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Abstract: ***Objective*** To evaluate the quality of the report about bone tumors in animal experiments. ***Methods*** The articles were searched about bone tumors in animal experiments at the four largest Chinese Journal database: CBM, CNKI, CBM, Wanfang from Jan. 2010 to Dec. 2012, and the language was Chinese. Each article was evaluated with ARRIVE guideline. Finally, the data acquired in the evaluation was statistically analysed with SPSS. ***Results*** There were 71 articles about bone tumors in animal experiments that met the criteria mentioned before. ARRIVE guideline was applied to evaluate those articles. Average score of the articles was 21.08±2.12. The qualities of the articles in the present work was generally middle or slightly low. ***Conclusion*** Generally, the quality of the report about bone tumor in animal experiment is quite undesirable with severe potential bias. In further studies, more attention should be paid to the design and report of animal experiments in order to guarantee the quality of the study.

Keywords: Bone tumor; Animal experiment; Report

动物实验是连接基础研究和临床试验的重要桥梁[1],动物研究报告存在不充分和不完整现象,阻碍动物实验被科学的利用[2]。有学者对胃癌、肺癌、神经系统动物实验报告质量进行评估,提示报告存在诸多不规范。为了促进动物实验报告质量进一步提高,国际实验动物 3R 中心(the national centre for the replacement, refinement and reduction of animals in research, NC3Rs)基于 CONSORT 声明[3]开发的动物实验研究报告指南(animal research: reporting: in vivo experiments guidelines, ARRIVE 指南),于 2010 年发表,包括题目、摘要、引言、方法、结果和讨论 6 部分,共 20 个条目及其细则[4-8]。

骨肿瘤是发生于骨骼系统的肿瘤,分为原发性骨肿瘤和转移性骨肿瘤,在我国原发性骨肿瘤的发生率为 2~3 人/10 万人,大约占全部肿瘤的 2%,男、女发病率之比为(1.5~2):1[9]。本研究旨在客观分析国内期刊发表骨肿瘤动物实验报告存在的问题,提出解决思路。

1 资料与方法

1.1 纳入与排除标准

1.1.1 纳入标准 1)研究对象:鼠(包括大鼠,小鼠,裸鼠等);2)研究疾病:骨肿瘤领域原始研究;3)

研究类型：动物实验。

1.1.2 排除标准 1)研究对象应用的是处死动物的组织；2)未涉及实验过程、技术和方法；3)综述类研究；4)基因类研究；5)没有报告任何研究方法信息的动物研究；6)骨转移癌类的研究。

1.2 文献检索

系统检索中国知网、中国生物医学文献数据库、维普及万方数据库。检索时间自2010年1月至2012年12月，检索文献语言为中文。主要检索词包括相关动物实验和研究领域的主要检索词：鼠、动物、体内，骨肿瘤、骨瘤、骨癌，动物实验、体内试验、在体试验、基础研究。

1.3 文献筛选

2位评价员(冯芹和冯小欢)独立阅读所获文献文题和摘要，在排除明显不符合纳入标准的研究后，对可能符合纳入标准的研究阅读全文，以确定是否真正符合纳入标准，并交叉核对纳入文献，对有分歧者通过讨论达成共识，必要时听取第3位研究者的意见。

1.4 资料提取

由2名评价员通过阅读全文，采用Excel 2003软件制定统一数据提取表提取数据，而后交叉核对，如遇分歧讨论解决或听取第3位研究者的意见。提取资料主要为研究的基本情况(如发表期刊、发表年限、研究设计类型、基金支持等方面)。

1.5 质量评价

2位评价员(冯芹和冯小欢)根据ARRIVE指南中各个条目独立评价纳入文献质量，包括题目、摘要、引言、方法、结果和讨论6部分，共20个条目及其近40项细则，评估内容涉及动物的数量和特点(包括种类、品系、雌雄和遗传背景)，饲养场所和饲养，所采用的实验方法，统计方法和分析方法(包括使用随机和盲法来减少偏倚)等，并对每个条目内容进行了简要解释，每一条目回答“是”为1分，回答“否”为0分，满分为39分。

1.6 数据处理及统计分析

所有数据录入特定Excel表格中，计算ARRIVE评价中的每一条目所得分和百分数以及每一篇文献的ARRIVE评价得分和百分数，采用SPSS16.0软件进行方差分析，计量资料以均数±标准差($\bar{x}\pm s$)表示，组间比较采用单因素方差分析(analysis of variance，ANOVA)，$P<0.05$为差异有统计学意义。

2 结果

2.1 文献检索结果

中国知网、中国生物医学文献数据库、维普、万方中最初共检选出2750篇文章，通过EndNote软件浏览题录和摘要后排除重复的、不相关的文章2564篇，剩余186篇，通过阅读全文进行复筛，排除因数据不全，综述类、基因类及转移瘤类的文献115篇，最终71篇文献被纳入。

2.2 纳入研究基本信息(表1)

3年纳入研究的篇数，研究是否为随机对照实验(RCT)及是否有基金支持的情况如下。

表1 纳入研究的基本信息(n，%)

类型	2010年	2011年	2012年	总计
纳入研究(篇)	25	23	23	71
RCT	25(100.00)	23(100.00)	19(82.61)	67(94.37)
有基金支持	18(72.00)	16(69.57)	15(65.22)	49(69.01)

2.3 纳入研究的报告质量评价(表2)

2.3.1 前言部分 背景内容部分没有一篇解释所用动物种类和模型的选择依据，阐述科学目的、适用范围，该研究与人体生物学的关联程度。

2.3.2 方法学部分 71篇研究中仅8篇(11.27%)提供了动物实验伦理审查的相关证书及与研究相关的国家或机构的动物护理和使用指南；实验设计部分，对于每个实验，给出简明扼要的研究设计详细信息。但没有一篇研究用时线图或流程图来解释复杂的研究设计是如何实施的；实验步骤部分，只有33篇(46.48%)交代了实验的具体地点(饲养笼，实验室，水迷宫)，没有一篇说明特定麻醉药的选择缘由及给药途径，药物剂量的选择原因；实验动物部分，仅有2篇(2.82%)提及实验动物的待遇，即实验前、中和后期相关动物福利评估和干预；只有1篇提到了独立重复实验(1.41%)；有15篇(21.13%)对实验动物进行了随机化分组，只有1篇(1.41%)在实验中使用了盲法；统计学处理部分，仅有5篇(7.04%)进行了假设检验，即描述用来评估数据是否符合统计学方法所要求的分布，及所采用的任何方法。

2.3.3 结果部分 仅14篇(19.72%)提供了完整的基线数据，即对于每个实验组，报告治疗或测试前动物的有关特征和健康状况(如体重、微生物状况和药物测试)，以表格形式表示；有13篇(18.31%)对实验失访动物进行了描述；没有一篇

文献提到结果中的不良反应。

表 2 纳入研究的 ARRIVE 评价报告信息

Item		Recommendation	Sum n(%) n=71	2010 n(%) n1=25	2011 n(%) n2=23	2012 n(%) n3=23
Title	1	Provide as accurate and concise a description of the content of the article as possible	70(98.59)	25(100.00)	22(95.65)	23(100.00)
Abstract	2	Provide as accurate summary of the background, research objectives(including details of the species or strain of animal used), key methods, principal findings, and conclusions of the study	70(98.59)	25(100.00)	22(95.65)	23(100.00)
Background	3a	a. Include sufficient scientific background(including relevant references to previous work)to understand the motivation and context for the study, and explain the experimental approach and rationale.	71(100.00)	23(100.00)	23(100.00)	23(100.00)
	3b	b. Explain how and why the animal species and model being used can address the scientific objectives and, where appropriate, the study's relevance to human biology	0(0.00)	0(0.00)	0(0.00)	0(0.00)
Objectives	4	Clearly describe the primary and any secondary objectives of the study, or specific hypotheses being tested	71(100.00)	25(100.00)	23(100.00)	23(100.00)
Ethical statement	5	Indicate the nature of the ethical review permissions relevant licences (eg. Animal [Scientific Procedures] Act 1986), and national or institutional guidelines for the care and use of animals, that cover the research.	8(11.27)	3(12.00)	3(13.04)	2(8.70)
Study design	6a	For each experiment, give brief details of the study design, including a. The number of experimental and control groups.	70(98.59)	25(100.00)	22(95.65)	23(100.00)
	6b	b; Any steps taken to minimize the effects of subjective bias when allocating animals to treatment(eg. randomization procedure)and when assessing results(eg. if done, describe who was blinded and when).	66(92.96)	25(100.00)	22(95.65)	19(82.61)
	6c	c. The experimental unit(eg. a single animal, group, or cage of animals).	71(100.00)	25(100.00)	23(100.00)	23(100.00)
	6d	d. A time-line diagram or flow chart can be useful to be illustrate how complex study designs were carried out.	0(0.00)	0(0.00)	0(0.00)	0(0.00)
Experimental procedures	7a	For each experiment and each experimental group, including controls, provide precise details of all procedures carried out. For example: a. How(eg. drug formulation and dose, site and route of administration, anaesthesia and analgesia used[including monitoring], surgical procedure, method of euthanasia). Provide details of any specialist equipment used, including suoolier(s).	71(100.00)	25(100.00)	23(100.00)	23(100.00)
	7b	b. When(eg. time of day)	64(90.14)	23(92.00)	18(78.26)	23(100.00)
	7c	c. Where(eg. home cage, laboratory, water maze).	33(46.48)	13(52.00)	12(52.17)	8(34.78)
	7d	d. Why(eg. rationale for choice of specific anaesthetic, route of administration, drug dose used).	0(0.00)	0(0.00)	0(0.00)	0(0.00)
Experimental animals	8a	a. Provide details of the animals used, including species, strain, sex, developmental stage(eg. mean or median age plus age range), and weight(eg. mean or median weight plus weight range).	70(98.59)	25(100.00)	22(95.65)	23(100.00)
	8b	b. Provide further relevant information such as the source of animals, international strain nomenclature, genetic modification status(eg. knock-out or transgenic), genotype, health/immune status, drug or test-naive, previous procedures, etc.	71(100.00)	25(100.00)	23(100.00)	23(100.00)

Housing and husbandry	9a	Provide details of: a. Housing(eg. type of facility, eg. specific pathogen free (SPF); type of cage or housing; bedding material; number of cage companions; tank shape and material etc. for fish).	43(60.56)	14(56.00)	19(82.61)	10(43.48)
	9b	b. Husbandry conditions(eg. breeding programme, light/dark cycle, temperature, quality of water etc. for fish, type of food, access to food and water, environmental enrichment).	50(70.42)	14(56.00)	17(73.91)	19(82.61)
	9c	c. Welfare-related assessments and interventions that were carried out before, during, or after the experiment.	2(2.82)	0(0.00)	0(0.00)	2(8.70)
Sample size	10a	a. Specify the total number of animals used in each experiment and the number of animals in each experimental group.	65(91.55)	25(100.00)	22(95.65)	18(78.26)
	10b	b. Explain how the number of animals was decided. Provide details of any sample size calculation used.	0(0.00)	0(0.00)	0(0.00)	0(0.00)
	10c	c. Indicate the number of independent replications of each experiment, if relevant.	1(1.41)	0(0.00)	0(0.00)	1(4.35)
Allocating animals to experimental groups	11a	a. Give full details of how animals were allocated to experimental groups, including randomization or matching if done.	15(21.13)	5(20.00)	3(13.04)	7(30.43)
	11b	b. Describe the order in which the animals in the different experimental groups were treated and assessed.	71(100.00)	25(100.00)	23(100.00)	23(100.00)
	11c		1(1.41)	1(4.00)	0(0.00)	0(0.00)
Experimental outcomes	12	Clearly define the primary and secondary experimental outcomes assessed(eg. ell death, molecular markers, behavioural changes).	71(100.00)	25(100.00)	23(100.00)	23(100.00)
Statistical methods	13a	a. Provide details of the statistical methods used for each analysis.	69(97.18)	24(96.00)	23(100.00)	22(95.65)
	13b	b. Specify the unit of analysis for each dataset(eg. single animal, single neuron).	66(92.96)	25(100.00)	22(95.65)	19(82.61)
	13c	c. Describe any methods used to assess whether the data met the assumptions of the statistical approach.	5(7.04)	1(4.00)	2(8.70)	2(8.70)
Baseline data	14	For each experimental group, report relevant characteristics and health status of animals(eg. weight, microbiological status, and drug-or test-naive) prior to treatment or testing(this information can often be tabulated).	14(19.72)	5(20.00)	0(0.00)	9(39.13)
Numbers analysed	15a	a. Report the number of animals in each group included in each analysis. Report absolute numbers(eg. 10/20, not 50%)	37(52.11)	12(48.00)	11(47.83)	14(60.87)
	15b	b. If any animals or data were not included in the analysis, explain why.	13(18.31)	5(20.00)	4(17.39)	4(17.39)
Outcomes and estimation	16	Report the results for each analysis carried out, with a measure of precision(eg. standard error or confidence interval).	68(95.77)	25(100.00)	22(95.65)	21(91.30)
Adverse events	17a	a. Give details of all important adverse events in each experimental group.	0(0.00)	00.00)	0(0.00)	0(0.00)
	17b	b. Describe any modifications to the experimental protocols made to reduce adverse events.	0(0.00)	0(0.00)	0(0.00)	0(0.00)
Interpretation/scientific implications	18a	a. Interpret the results, taking into account the study objectives and hypotheses, current theory and other relevant studies in the literature.	71(100.00)	25(100.00)	23(100.00)	23(100.00)
	18b	b. Comment on the study limitations including any potential sources of bias, any limitations of the animal model, and the imprecision associated with the results[2].	12(16.90)	6(24.00)	5(21.74)	1(4.35)
	18c	c. Describe any implications of your experimental methods or findings for the replacement, refinement or reduction(the 3Rs)of the use of animals in research.	0(0.00)	0(0.00)	0(0.00)	0(0.00)
Generalisability/translation	19	Comment on Whether, and how, the findings of this study are likely to translate to other species or systems, including any relevance to human biology.	15(21.13)	6(24.00)	9(39.13)	0(0.00)
统计			21.08±2.12	21.28±1.84	21.13±2.01	20.83±2.55

2.3.4 结论部分 只有 12 篇(16.9%)在结论中评价了研究的局限性,包括任何潜在的偏倚来源,动物模型的局限性以及与结果相关的不精确性;没有一篇描述该研究方法或研究发现对于替代、优化或减少动物使用(3R 原则)的意义;仅有 15 篇(21.13%)评论了是否或如何使本研究成果转化到其他物种或系统,包括与人体生物学相关的研究。

3 讨论

在我国,原发性骨与软组织肿瘤约占全身肿瘤的 2%~3%,其中 1/3 是恶性骨肿瘤[10],尤其骨肉瘤由于其恶性程度高,转移发生早,因而其致残率及死亡率位于骨肿瘤之首[11]。动物实验仍是骨肿瘤研究能够应用于临床的重要桥梁。

国外已有针对动物实验报告质量的评估报告[12-16],结果提示:大多数生物期刊未能完整提供如何描述动物研究的信息。ARRIVE 指南基于 CONSORT 声明[17]制定。本研究根据 ARRIVE 指南中各个条目独立评价纳入文献质量。本文共收集 2010 年 1 月至 2012 年 12 月中文期刊发表的骨肿瘤动物实验的研究 71 篇。结果显示:从发表时间看,3 年间的 ARRIVE 评价得分差异无统计学意义,骨肿瘤动物实验的报告质量普遍中等偏低,尤其在方法学部分,进行动物实验的相关操作过程没有具体详细地说明,如实验的随机化分组及盲法的应用是减少偏倚,提高实验结果有效性的关键因素。本研究中只有 15 篇(21.13%)对实验动物进行了随机化分组,仅有 1 篇(1.41%)在实验中使用了盲法;还有实验动物的失访情况及不良反应,对整篇研究的最终结果也会产生一定影响,而研究发现仅有 13 篇(18.31%)对实验失访动物进行了描述;没有一篇文献提到结果中的不良反应;同时我们还发现动物实验研究的统计学分析不够严谨,仅有 5 篇(7.04%)进行了假设检验。

本研究存在以下几点局限性,如样本量不够大,样本来源范围有限以及评价员进行 ARRIVE 评价时存在一定的主观偏倚等。

综上所述,我国骨肿瘤的动物实验研究报告质量总体中等偏低。研究者们应遵循 ARRIVE 指南的标准,提高动物实验的设计、实施、报告质量以及对动物实验的鉴别能力,评估水平,这样骨肿瘤动物实验的研究成果才能有效地应用于临床,从而推动骨肿瘤研究更进一步地深入。

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