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Fischer-344 rats are unsuitable for the MCAO filament model due to their cerebrovascular anatomy

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Abstract

Middle cerebral artery occlusion (MCAO) in Fischer-344 rats results in a small variance of infarct size. However, complications are frequent especially in aged Fisher-344 rats undergoing endovascular suture occlusion of the middle cerebral artery. Analyzing our experiences with 165 Wistar, 13 Sprague-Dawley and 10 F-344 rats, we compared the incidence of impossible thread advancement and subarachnoid hemorrhage, respectively. Magnetic resonance angiography (MRA) was applied to study the course of the internal carotid artery (ICA) in Fischer and Wistar rats. Finally, we performed a structured review of the literature from 1991 to 2005 evaluating reports on Fischer rats subjected to intraluminal filament MCAO. Complications like fruitless filament advancement or subarachnoid hemorrhage were found to be significantly more frequent in Fischer rats than in other strains. MRA revealed significantly more pronounced kinking of the ICA in F-344 than in Wistar rats. In seven publications available on filament MCAO in F-344 rats, complication rates of 50–100% were reported, corroborating our data. Surgical difficulties accompanied by high complication rates due to their cerebrovascular anatomy make Fischer rats unsuitable for filament MCAO. If the use of Fischer rats for studies on focal cerebral ischemia is indicated, other ischemia models than intraluminal suture occlusion should be chosen.

Keywords: Cerebral ischemia; Focal; Disease models; Animal; Rats; Middle cerebral artery occlusion; Carotid artery; Internal

1. Introduction

The endovascular filament model of middle cerebral artery occlusion (MCAO) is a frequently applied model of focal cerebral ischemia. In contrast to outbred strains like Wistar or Sprague-Dawley (SD), inbred Fischer F-344 rats have been reported to develop more reproducible brain infarcts after MCAO (Duverger and MacKenzie, 1988; Herz et al., 1996b). In addition, experimental studies involving homologous cell or tissue transplantation favor the use of inbred rats like the Fischer-344 strain to avoid graft rejection. According to the literature, aged Fischer rats weighing 350–450 g are not suitable for the intraluminal suture MCAO due to difficulties in advancing the filament. The exact cause of the phenomenon is yet undetermined, but was attributed to the age of the animals (Wang et al., 1995). In order to clarify whether young adult Fischer rats can be successfully used for the filament stroke model, we performed a systematic literature enquiry on the use of Fischer rats for filament MCAO and evaluated our own experimental data on the success rates of that model in different strains. We further compared the anatomy of the internal carotid artery (ICA) in F-344 rats to that of the outbred Wistar strain to elucidate the cause for difficulties in advancing the filament.

2. Materials and methods

2.1. Literature review

A systematic literature search was performed on the SilverPlatter Medline database browsed by Ovid Technologies ERL WebSPIRS 5.11 in February 2006, limited to the publication years 1991–2005. In a first step, publications involving transient MCAO were identified by applying the Boolean expression

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"((rat OR rats) AND (mca?o OR (middle NEAR cerebral NEAR artery NEAR occlusion) OR (mca NEAR occlusion)) NOT ((mca?o OR (middle NEAR cerebral NEAR artery NEAR occlusion) OR (mca NEAR occlusion)) NEAR permanent))". In a second step, the proportion of different strains was evaluated. Fischer rats were identified by combining the first search with the string "AND (((fischer OR fisher OR f?344) IN AB) OR ((fischer OR fisher OR f?344) IN TI) OR (EXPLODE "Rats-Inbred-F344" IN MIME,MJME))", Wistar rats were searched for with "AND ((explode "Rats-Wistar"/all SUBHEADINGS in MIME,MJME,PT) OR (Wistar*))", Long-Evans rats with "AND ((long evans) OR (long?evans) OR (explode "Rats-Long-Evans"/all SUBHEADINGS in MIME,MJME,PT))", spontaneously hypertensive rats (SHR) by "AND ((spont* NEAR hypertensi*) OR (explode "Rats-Inbred-SHR"/all SUBHEADINGS in MIME,MJME,PT))" and inbred strains other than Fischer rats or SHR by exploding all inbred strains except F-344 rats and SHR via the thesaurus.

Concerning publications on Fischer rats, reports without specific mention of the filament MCAO model in the methods section were excluded. Remaining papers were reviewed for information on success rate, complications and the application of laser Doppler flow monitoring (LDF) to monitor MCAO. Corresponding authors of larger studies lacking important information on complications and success rates were contacted by email and asked for additional information on this topic.

2.2. Animal experiments

Animal care and all experimental procedures were conducted in accordance with German laws governing animal care and with the European Communities Council Directive (86/609/EEC). Protocols were approved by the Ethics Committee for Animal Research of the local authorities.

Male Wistar (Crl:WI, $n = 165$) and SD (Crl:CD(SD), $n = 13$) rats were purchased from Charles River Laboratories (Sulzfeld, Germany) and Fischer F-344 rats from Charles River (F-344/DuCrl, $n = 6$) or Harlan Winkelmann (Borchen, Germany; F344/NHsd, $n = 4$). Animal characteristics are displayed in Table 1.

2.3. MCAO procedure

Occlusion of the middle cerebral artery (MCA) was performed according to Longa (Longa et al., 1989) with modifications herein described. Anesthesia was performed with 1.5–2% isoflurane in N_2O/O_2 (70/30%) under endotracheal intubation and mechanical ventilation. The bregma was exposed and the skull bone was countersunk at two 3 mm × 3 mm areas over both MCA territories of supply for bilateral monitoring of local cortical blood flow by LDF (MBF3D, Moor Instruments, Wilmington, Delaware). Subsequently, the right external carotid artery was transected for filament introduction. A silicone-coated 4-0 monofilament with a final diameter of 0.28 mm was gently advanced through the ICA until its tip occluded the origin of the MCA. In case of difficulties during filament advancement, attempts with thinner filaments (6-0, 8-0) and alternative head positions were conducted. Reperfusion was initiated by filament withdrawal after 90 min of ischemia. Successful MCAO was defined as uncomplicated filament introduction leading to a steep, sustained ipsilateral LDF decline without serious complications (filament introduction impossible, SAH or intraprocedural death). SAH was detected by irreversible bilateral LDF drop (Schmid-Elsaesser et al., 1998).

2.4. MR angiography

For magnetic resonance imaging (MRI), anesthesia was performed using subcutaneous injections of xylazine (4–8 mg/kg body weight) and ketamine (60–120 mg/kg) as described elsewhere (Dittmar et al., 2004). Magnetic resonance angiography (MRA) was performed on three naive Fischer rats weighing 230–336 g and three naive Wistar rats weighing 276–285 g with a 1.5T clinical MRI scanner (Magnetom Sonata, Siemens, Erlangen, Germany) as described elsewhere (Fehm et al., 2005). The rat's skull was positioned in the small loop coil for signal detection. Fast scout scans were used for positioning of the three-dimensional time-of-flight (TOF) angiography slaps with the following imaging parameters: repetition time 47 ms, echo time 7.52 ms, flip angle 25°, field of view 85 mm, matrix dimension 384 × 384, 2 slaps, 30 slices, slice thickness 0.5 mm, acquisition time 11:35 min. Reconstructions of angiograms were obtained by generating maximum intensity projections using the standard MRI software (syngo MR 2002, B).

2.5. Evaluation of ICA kinking

To compare vessel kinking, on transverse and sagittal MRA images the ICA course was divided into straight sections. Through these sections, lines were drawn and the angles between

Table 1
Animal characteristics and complication rates

Species	*n*	Weight (g)	Mean age[a] (weeks)	Complications during filament introduction (*n*/%)		
				SAH	No filament forwarding	None
Wistar	165	270 ± 22	9–10	8/4.8	1/0.6	156/94.5
Sprague-Dawley	13	335 ± 60	9–10	0/0.0	0/0.0	13/100.0
Fischer F-344	10	244 ± 19	12–13	5/50.0	4/40.0	1/10.0

Body weight (mean ± standard deviation), age, and rates of complications during suture introduction in different strains in the MCAO filament model. Fischer rats differ in a statistically significant manner from other strains ($P \leq 0.012$) concerning weight and frequency of complications.

[a] Age was calculated from mean body weight according to Charles River growth diagrams. SAH = subarachnoid hemorrhage.