

Forced Needle Advancement During Needle-Nerve Contact in a Porcine Model: Histological Outcome

Thorsten Steinfeldt, MD,* Sabine Poeschl,* Wilhelm Nimphius, MD,† Juergen Graf, MD,*‡ Martin Zoremba, MD,* Hans-Helge Mueller, PhD,§ Hinnerk Wulf, MD,* and Frank Dette, MD*

BACKGROUND: In this study, we determined whether needle advancement during needle-nerve contact (forced needle-nerve contact) is associated with a higher risk of nerve injury compared with needle-nerve contact without needle advancement (nonforced needle-nerve contact).

METHODS: In 8 anesthetized pigs, the brachial plexus nerves underwent forced (0.15 Newton) or nonforced (0.0 Newton) needle-nerve contact without nerve penetration. The grade of nerve injury was histologically assessed using an objective score ranging from 0 (no injury) to 4 (severe injury).

RESULTS: Sixty-nine nerves, including controls, were examined. Histology revealed a significant difference between forced and nonforced needle-nerve contact (median [interquartile range] 3 [2–4] vs 2 [1–2]; $P = 0.004$). Myelin damage and intraneural hematoma occurred only after forced needle-nerve contact.

CONCLUSIONS: The severity of structural nerve injury after needle-nerve contact was directly related to force exposure via needle advancement. (Anesth Analg 2011;113:417–20)

During needle advancement during peripheral block, nerve penetration is typically avoided to reduce the likelihood of nerve injury.¹ Indeed, even close needle-nerve proximity with needle-nerve contact (but no penetration) is frequently denoted a traumatic approach.² However, there is a lack of data regarding the histological outcome after needle-nerve contact without penetration. Specifically, nerve strain and pressure particularly induced by forced needle advancement have not been evaluated.

The aim of this animal study was to objectively assess the risk and severity of needle-related nerve injury after forced needle advancement in contrast to nerves exposed to nonforced needle-nerve contact only.

METHODS

After IRB approval (Ref: 50/2007 No. 2, 50/2009 No. 2; Regional Board, Giessen, Hessen, Germany), we studied 8 pigs (Deutsche Landrasse) weighing 27 to 39 kg. Under general anesthesia, both axillary regions were opened by blunt dissection as previously reported.^{3–5}

The median, radial, and axillary nerves underwent needle placement bilaterally with large-sized needles as frequently used for regional catheter techniques (StimuCath™, Tuohy 18 gauge, 8 cm in length; Arrow, Erding, Germany). The needles with a connected force gauge

From the *Department of Anaesthesiology and Intensive Care Therapy, and †Institute of Pathology, Philipps University Marburg, Marburg; ‡Aero Medical Center, Medizinischer Dienst, Lufthansa AG, Frankfurt am Main; and §Department of Medical Informatics, Biostatistics and Epidemiology, Ludwig-Maximilians-University, Munich, Germany.

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Address correspondence to Thorsten Steinfeldt, MD, Philipps University, Department of Anaesthesiology and Intensive Care Therapy, University Hospital Giessen-Marburg, Campus Marburg, Baldingerstrasse, 35032 Marburg, Germany. Address e-mail to steinfeldt@gmx.de.

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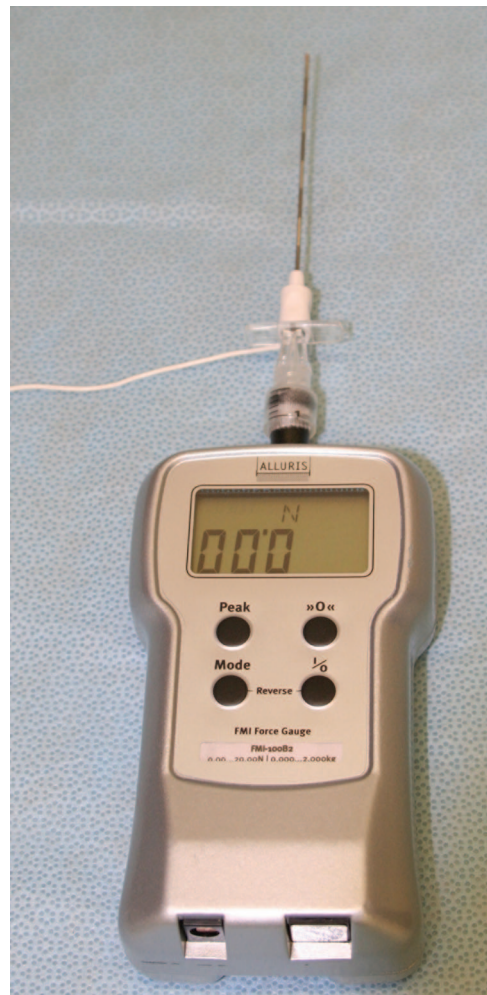


Figure 1. A digital force gauge instrument (FMI 100B2®; Alluris GmbH, Freiburg, Germany) was connected to a needle (StimuCath™, Tuohy 18 gauge, 8 cm in length; Arrow, Erding, Germany) via a Luer lock joint. The time interval between 2 measured points for force measurement was 0.2 second during needle placement. Forces were quantified in Newton.

Table 1. Number of Nerve Specimens and Histological Slices, and Occurrence of Intraneural Hematoma and Myelin/Fascicle Damage in Injury Groups and Controls

	Nonforced NNC (0.0 N)	Forced NNC (0.15 N)	Controls		
			No treatment (brachial plexus)	No treatment (sciatic nerve)	Positive control (nerve penetration)
Nerve specimen (n)	23	22	8	8	8
Histological slices (n) HE/CD68/KB	3050/62/94	2974/56/106	480/16/16	420/16/16	580/16/16
Hematoma (HE), n, specimen (%)	0 (0%)	10 (45%)	0	0	5 (63%)
Avital myelin (KB), n, specimen (%)	0 (0%)	7 (32%)	0	0	3 (38%)
Monocytic cells, mean ± SD (%)	39 ± 9	44 ± 10	—	—	38 ± 7

HE = hematoxylin and eosin staining; CD68 = specific staining of CD68-positive leucocytes (macrophages) by application of an immunohistochemistry method⁸; KB = myelin staining according to the Klüver-Barrera method⁹; NNC = needle-nerve contact.

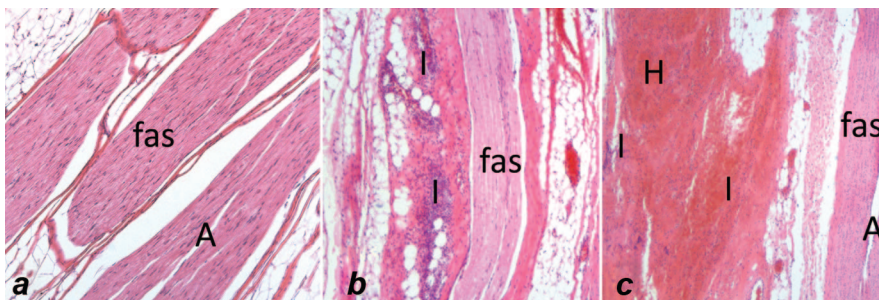


Figure 2. a, Sciatic nerve (negative control). Longitudinal microscopic view. Score value, 0. b, Histological changes in the radial nerve after needle-nerve contact (0.0 N). Longitudinal microscopic view. Distinctive signs of post-traumatic inflammation. Score value, 2. c, Histological changes in the median nerve after forced needle-nerve contact (0.15 N). Tangential microscopic view. Score value, 4. Original magnification ×100, hematoxylin and eosin staining. Fas = fascicle of nerve; I = inflammatory cells; H = hematoma; A = artifact.

instrument (Fig. 1) were placed perpendicular to the target nerves. For nonforced needle placement, the needles were sited without pressure on the surface of the nerve only. In this position, no force (0.0 Newton [N]) was measured during needle-nerve contact. For forced needle placement, the needles were slowly advanced on the nerve's surface until a predefined force of 0.15 N was measured. The needles were left in this position for 40 seconds based on the assumption that in clinical practice it takes approximately 40 seconds from final needle positioning to the completion of the local anesthetic injection and subsequent needle retraction. The force gauge instrument and the connected needle were guided manually. If the adjusted target force (0.15 N) declined during needle position maintenance, the needle location was adjusted until the predefined force was restored. The selection of force intensity was based on 100 pilot force measurements for needle advancement in porcine muscular tissue, where a minimal force of 0.16 ± 0.07 N (mean ± SD) was required to push the needle through the tissue.

After 40 seconds, the needles were retracted carefully. As a positive control for maximum needle trauma, needle-nerve penetration was performed at the left tibial nerve. After completion of the interventions, the surgical wounds were closed. Cefuroxime $80 \text{ mg} \cdot \text{kg}^{-1} \cdot \text{d}^{-1}$ IV was administered and anesthesia was maintained. After 48 hours, the brachial plexus, the right caudal pectoral nerve (negative control of brachial plexus), the left tibial (positive control), and the left sciatic nerve (negative control for systemic confounders) were extracted and the animals were euthanized. The specimens were processed for visual examination and the detection of trauma-related inflammation^{6,7} (hematoxylin and eosin, i.e., CD68-immunohistochemistry⁸ for visualization of macrophages), myelin damage (Klüver-Barrera staining⁹), and intraneural hematoma. The grade of nerve injury was scored ranging from 0 (no injury) to 4 (severe injury) as

reported recently.³⁻⁵ The primary outcome measure was the nerve damage according to a grading after needle-nerve contact with or without needle advancement.

Data were analyzed using the nonparametric Wilcoxon-Mann-Whitney test. A power analysis with a power of 0.8 ($1 - \beta$) and $\alpha \leq 0.05$ indicated that a sample size of at least 20 specimens for each group (forced and nonforced needle-nerve contact) would be required to detect a score value difference of 1.0. Statistical analyses were performed using SPSS software for Windows (Release 15.0; SPSS, Inc., Chicago, IL).

Data are presented as median with 25th and 75th percentiles (interquartile range), mean (SD), or frequencies as appropriate. Nonparametric data were analyzed using Wilcoxon-Mann-Whitney test. Level of statistical significance was taken at $P \leq 0.05$.

RESULTS

A total of 69 nerves were evaluated (Table 1). Signs of posttraumatic inflammation (Fig. 2) were found in all except the negative controls. The ratio of monocytic cells to leukocytes was increased corresponding to trauma-related inflammation (Table 1, Fig. 3). In contrast to the negative control (noninjury groups: sciatic nerve, brachial plexus), the positive control (nerve penetration) showed signs of significant nerve injury (Table 1).

After forced needle-nerve contact and nerve penetration (positive control), hematoma (Fig. 2) and myelin damage (Fig. 4) were observed (Table 1), associated with a considerable accumulation of inflammatory cells (Fig. 2, Table 1). In contrast, inflammation without any structural alterations was found after nonforced needle-nerve contact (Fig. 2). The median score value for nerve injury was significantly higher (median [interquartile range] 3 [2-4]) after forced needle-nerve contact compared with nonforced needle-nerve contact (2 [1-2]) ($P = 0.004$).

DISCUSSION

Force exposure on peripheral nerves (force, 0.15 N) caused by needle advancement during needle-nerve contact led to significant structural nerve injury compared with nonforced nerve contact. We used an objective score including several aspects of histological signs of nerve injury and posttraumatic inflammation representing elapsed nerve trauma.³⁻⁵ The results of our control groups emphasize both the validity of the experimental intervention and the construct validity of the applied score³⁻⁵: a maximum trauma and a nontraumatic intervention were easily distinguishable and reproducible within the categories of the score.

This investigation has a number of limitations. First, there are significant differences between humans and pigs that limit the ability to generalize our data. Second, we lacked any functional assessment of nerve function. However, behavioral assessment is challenging, especially in larger animals such as pigs. Third, in our experimental setting, we used an "open brachial plexus model." Percutaneous placement of needles might have been desirable for a variety of reasons (e.g., closer to clinical practice, abdication of surgery). Thus, the challenge to execute and subsequently identify the site of needle-nerve contact is an

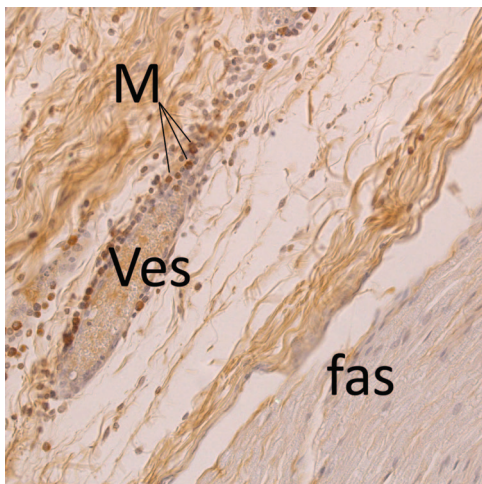
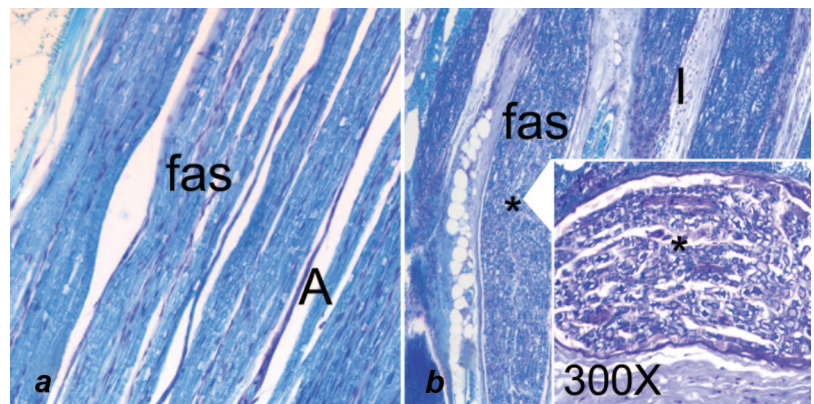


Figure 3. Histological changes in the axillary nerve after nonforced needle-nerve contact (0.0 N). Tangential microscopic view ($\times 400$, immunohistochemistry⁸ for specific staining of CD68-positive macrophages). Score value, 2. M = macrophages; fas = fascicle of nerve.

Figure 4. a, Unaffected myelin of the caudal pectoral nerve (negative control). Tangential microscopic view ($\times 200$, staining according to Klüver-Barrera⁹). Score value, 0. b, Histological changes in the median nerve after forced needle-nerve contact (0.15 N). Longitudinal microscopic view ($\times 100$, staining according to Klüver-Barrera⁹). *Demyelination: myelin appears inconsistent, stretched, swollen, and lower stained; inset (300 \times) depicts tangential microscopic view. Score value, 4. Fas = fascicle; A = artifact; I = inflammatory cells.



obstacle for a controlled and reproducible experimental setting.

In conclusion, this study demonstrates that needle-nerve contact together with forced needle advancement in pigs may lead to severe structural nerve injury. In contrast, no structural impairment was detectable after needle-nerve contact without force exposure. ■■

DISCLOSURES

Name: Thorsten Steinfeldt, MD.

Contribution: This author helped design the study, conduct the study, analyze the data, and write the manuscript.

Attestation: Thorsten Steinfeldt has seen the original study data, reviewed the analysis of the data, approved the final manuscript, and is the author responsible for archiving the study files.

Name: Sabine Poeschl.

Contribution: This author helped conduct the study.

Attestation: Sabine Poeschl has seen the original study data, reviewed the analysis of the data, and approved the final manuscript.

Name: Wilhelm Nimphius, MD.

Contribution: This author helped conduct the study and analyze the data.

Attestation: Wilhelm Nimphius has seen the original study data, reviewed the analysis of the data, and approved the final manuscript.

Name: Juergen Graf, MD.

Contribution: This author helped design the study and write the manuscript.

Attestation: Juergen Graf has seen the original study data, reviewed the analysis of the data, and approved the final manuscript.

Name: Martin Zoremba, MD.

Contribution: This author helped conduct the study.

Attestation: Martin Zoremba has seen the original study data, reviewed the analysis of the data, and approved the final manuscript.

Name: Hans-Helge Mueller, PhD.

Contribution: This author helped design the study and analyze the data.

Attestation: Hans-Helge Mueller has seen the original study data, reviewed the analysis of the data, and approved the final manuscript.

Name: Hinnerk Wulf, MD.

Contribution: This author helped write the manuscript.

Attestation: Hinnerk Wulf has seen the original study data, reviewed the analysis of the data, and approved the final manuscript.

Name: Frank Dette, MD.

Contribution: This author helped design the study, conduct the study, analyze the data, and write the manuscript.

Attestation: Frank Dette has seen the original study data, reviewed the analysis of the data, and approved the final manuscript.

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REFERENCES

1. Neal JM. Ultrasound-guided regional anesthesia and patient safety: an evidence-based analysis. *Reg Anesth Pain Med* 2010;35:S59–67
2. Neal JM, Gerancher JC, Hebl JR, Ilfeld BM, McCartney CJ, Franco CD, Hogan QH. Upper extremity regional anesthesia: essentials of our current understanding, 2008. *Reg Anesth Pain Med* 2009;34:134–70
3. Steinfeldt T, Nimphius W, Werner T, Vassiliou T, Kill C, Karakas E, Wulf H, Graf J. Nerve injury by needle nerve perforation in regional anaesthesia: does size matter? *Br J Anaesth* 2010;104:245–53
4. Steinfeldt T, Nimphius W, Wurps M, Eberhart L, Vassiliou T, Kill C, Wulf H, Graf J. Nerve perforation with pencil point or short bevelled needles: histological outcome. *Acta Anaesthesiol Scand* 2010;54:993–9
5. Steinfeldt T, Werner T, Nimphius W, Wiesmann T, Kill C, Muller HH, Wulf H, Graf J. Histological analysis after peripheral nerve puncture with pencil-point or Tuohy needle tip. *Anesth Analg* 2010;112:465–70
6. Mueller M, Wacker K, Ringelstein EB, Hickey WF, Imai Y, Kiefer R. Rapid response of identified resident endoneurial macrophages to nerve injury. *Am J Pathol* 2001;159:2187–97
7. Mueller M, Leonhard C, Wacker K, Ringelstein EB, Okabe M, Hickey WF, Kiefer R. Macrophage response to peripheral nerve injury: the quantitative contribution of resident and hematogenous macrophages. *Lab Invest* 2003;83:175–85
8. Falini B, Flenghi L, Pileri S, Gambacorta M, Bigerna B, Durkop H, Eitelbach F, Thiele J, Pacini R, Cavaliere A. PG-M1: a new monoclonal antibody directed against a fixative-resistant epitope on the macrophage-restricted form of the CD68 molecule. *Am J Pathol* 1993;142:1359–72
9. Klüver H, Barrera E. A method for the combined staining of cells and fibers in the nervous system. *J Neuropathol Exp Neurol* 1953;12:400–3