through transcription factor NFAT. J. Neurochem. 108, 115-25 (2009)

3) Tsuda M *, Toyomitsu E *, Kometani M, Tozaki-Saitoh H, Inoue K, *equally contributed. Mechanisms underlying fibronectin-induced upregulation of P2X4R expression in microglia: distinct roles of PI3K-Akt and MEK-ERK signaling pathways. Journal of Cellular and Molecular Medicine. 2009 Feb 27. [Epub ahead of print]

4) Shinozaki Y, Sumitomo K, Tsuda M, Koizumi S, Inoue K, Torimitsu K, Direct observation of ATP-induced conformational changes in single P2X4 receptors. PLoS Biology. 5;7(5):e103 (2009) [Epub ahead of print]

5) Tsuda M*, Masuda T*, Kitano J, Shimoyama H, Tozaki-Saitoh H, Inoue K, *equally contributed. IFN-γ receptor signaling mediates spinal microglia activation driving neuropathic pain. Pro. Natl. Acad. Sci. USA. 106, 8032-8037 (2009)

6) Nagata K*, Imai T*, Yamashita T, Tozaki-Saitoh H, Tsuda M, Inoue K, *equally contributed. Antidepressants inhibit P2X4 receptor function: a possible involvement in neuropathic pain relief. Mol Pain. 5:20 (2009)

7) Hasegawa S, Kohro Y, Tsuda M, Inoue K, Activation of cytosolic phospholipase A2 in dorsal root ganglion neurons by Ca2+/calmodulin-dependent protein kinase II after peripheral nerve injury. Mol Pain. 5:22 (2009)

8) Masuda J, Tsuda M, Tozaki-Saitoh H, Inoue K, Intrathecal delivery of PDGF produces tactile allodynia through its receptors in spinal microglia. Mol Pain. 5:23 (2009)

9) Ohsawa K, Irino Y, Sanagi T, Nakamura Y, Suzuki E, Inoue K, Kohsaka S. P2Y12 receptor-mediated integrin-betal activation regulates microglial process extension induced by ATP. Glia. 2010 May;58(7):790-801.

10) S. Hasegawa, Y. Kohro, M. Shiratori, S. Ishii, T. Shimizu, M. Tsuda and K. Inoue. Role of PAF Receptor in Proinflammatory Cytokine Expression in the Dorsal Root Ganglion and Tactile Allodynia in a Rodent Model of Neuropathic Pain. PLoS ONE 2010 May 3;5(5):e10467.

11) M. Shiratori*, H. Tozaki-Saitoh*, M. Yoshitake, M. Tsuda, K. Inoue. P2X7 receptor activation induces CXCL2 production in microglia through NFAT and PKC/MAPK pathways. J Neurochem., in press

12) M. Maeda, M. Tsuda, H. Tozaki-Saitoh, K. Inoue, H. Kiyama. Nerve injury-activated microglia engulf myelinated axons in a P2Y12 signaling-dependent manner in the dorsal horn. Glia, in press

13) N. Kusunose, S. Koyanagi , K. Hamamura, N. Matsunaga, M. Yoshida, T. Uchida, M. Tsuda, K. Inoue and S. Ohdo. Molecular basis for the dosing time-dependency of anti-allodynic effects of gabapentin in a mouse model of neuropathic pain. Molecular Pain 2010, 6:83 (doi:10.1186/1744-8069-6-83)

14) M. Tsuda, Y. Kohro, T. Yano, T. Tsujikawa, J. Kitano, H. Tozaki-Saitoh, S. Koyanagi, S. Ohdo, R-R Ji, M. W. Salter, K. Inoue. JAK-STAT3 pathway regulates spinal astrocyte proliferation and neuropathic pain maintenance in rats. Brain 134(4): 1127-1139, 2011

15) Biber K, Tsuda M, Saitoh-Tozaki H, Tsukamoto K, Toyomitsu E, Masuda T, Boddeke H, Inoue K. Neuronal CCL21 up-regulates microglia P2X4 expression and initiates neuropathic pain development. EMBO J 30:1864-73, 2011

16) M. Nishida, M. Ogushi, R. Suda, M. Toyotaka, S. Saiki, N. Kitajima, M. Nakaya, K.-M. Kim, T. Ide, Y. Sato, K. Inoue, and H. Kurose. Heterologous down-regulation of angiotensin type 1 receptors by purinergic P2Y2 receptor stimulation through S-nitrosylation of NF-κB, PNAS 108(16):6662-7, 2011

17) K. Kuboyama, H. Harada, H. Tozaki-Saitoh, M. Tsuda, K. Ushijima and K. Inoue.

Astrocytic P2Y1 receptor is involved in the regulation of cytokine/chemokine transcription and cerebral damage in a rat model of cerebral ischemia. J Cereb Blood Flow Metab. 31(9):1930-41, 2011 (表紙を飾る)

18) Kataoka A, Koga Y, Uesugi A, Tozaki-Saitoh H, Tsuda M, Inoue K. Involvement of vasodilator-stimulated phosphoprotein in UDP-induced microglial actin aggregation via PKC- and Rho-dependent pathways. Purinergic Signal. 2011 May 13. [Epub ahead of print]

19) K. Kuboyama, M. Tsuda, M. Tsutsui, Y. Toyohara, H. Tozaki-Saitoh, H. Shimokawa, N. Yanagihara and K. Inoue. Reduced spinal microglial activation and neuropathic pain after nerve injury in mice lacking all three nitric oxide synthases. Mol Pain 7:50 2011

20) Y. Hayashi, K. Kawaji, L. Sun, X. Zhang, K. Koyano, T. Yokoyama, S. Kohsaka, K. Inoue, and H. Nakanishi. Microglial Ca2+-activated K+ Channels Are Possible Molecular Targets for the Analgesic Effects of S-ketamine on Neuropathic Pain. J. Neurosci 31(48):17370-82, 2011

21) Toyomitsu E, Tsuda M, Yamashita T, Tozaki-Saitoh H, Tanaka Y, Inoue K. CCL2 promotes P2X4 receptor trafficking to the cell surface of microglia. Purinergic Signal. 2012 Jan 6. [Epub ahead of print]

22) Tozaki-Saitoh H, Tsuda M, Inoue K. Role of purinergic receptors in CNS function and neuroprotection. Adv Pharmacol. 2011;61:495-528.

23) Tsuda M, Tozaki-Saitoh H, Inoue K. Platelet-activating factor and pain. Biol Pharm Bull. 2011;34(8):1159-62.

24) Inoue K, Tsuda M. Purinergic systems, neuropathic pain and the role of microglia. Exp Neurol. 2011 Sep 17. [Epub ahead of print]

25) Tsuda M, Tozaki-Saitoh H, Inoue K. Purinergic system, microglia and neuropathic pain. Curr Opin Pharmacol. 2011 Oct 27. [Epub ahead of print]

２．学会発表

1) 津田誠、井上和秀。ミクログリアと疼痛。52回日本神経化学会大会・シンポジウム、(2009.6 伊香保)

2) Koga Y, Kataoka A, Tozaki-Saitoh H, Tsuda M, Inoue K, Possible involvement of VASP phosphorylation in the P2Y6 receptor-induced phagocytosis of microglia. Fukuoka Purine 2009: International Symposium on Purinergic Signalling in New Strategy of Drug Discovery; a satellite symposium for the 36th Congress of the International Union of Physiological Sciences, (2009.7 Fukuoka)

3) Yano T, Tsuda M, Tsujikawa T, Kohro Y, Inoue K, Activation of JAK2/STAT3 signaling pathway in spinal cord astrocytes induces neuropathic pain after peripheral nerve injury. Fukuoka Purine 2009: International Symposium on Purinergic Signalling in New Strategy of Drug Discovery; a satellite symposium for the 36th Congress of the International Union of Physiological Sciences, (2009.7 Fukuoka)

4) Yamashita T, Tsuda M, Tozaki-Saitoh H, Inoue K. The possible effect on surface membrane P2X4 receptor expression in microglia by inhibition of V-ATPase activity. Fukuoka Purine 2009: International Symposium on Purinergic Signalling in New Strategy of Drug Discovery; a satellite symposium for the 36th Congress of the International Union of Physiological Sciences, (2009.7 Fukuoka)

5) Kuboyama K, Tsuda M, Naraki Y, Tsutsui M, Toyohara Y, Tozaki-Saitoh H, Shimokawa H, Yanagihara N, Inoue K, Spinal microglial activation and tactile allodynia after nerve injury are regulated by nitric oxide synthase. Fukuoka Purine 2009: International Symposium on Purinergic Signalling in New Strategy of Drug Discovery; a satellite symposium for the 36th Congress of the International Union of Physiological Sciences, (2009.7 Fukuoka)

6) Fujishita K, Nakao A, Inoue K, Koizumi S, Mechanism underlying upregulation of P2Y6 receptors in microglia in kainate-induced injury model. Fukuoka Purine 2009: International Symposium on Purinergic Signalling in New Strategy of Drug Discovery; a satellite symposium for the 36th Congress of the International Union of Physiological Sciences, (2009.7 Fukuoka)

7) Kometani M, Matsumura Y, Nagata K, Tsuda M, Inoue K, Analgesic action of clomipramine and maprotiline via P2X4 receptor inhibition. Fukuoka Purine 2009: International Symposium on Purinergic Signalling in New Strategy of Drug Discovery; a satellite symposium for the 36th Congress of the International Union of Physiological Sciences, (2009.7 Fukuoka)

8) Toyomitsu E, Tsuda M, Inoue K. Fibronectin increases P2X4R protein expression on the surface of microglial cells. Fukuoka Purine 2009: International Symposium on Purinergic Signalling in New Strategy of Drug Discovery; a satellite symposium for the 36th Congress of the International Union of Physiological Sciences, (2009.7 Fukuoka)

9) Shimoyama H, Tsukamoto K, Tozaki-Saitoh H, Tsuda M, Inoue K, Mechanisms of remitting neuropathic pain by activation of CB2 receptors. Fukuoka Purine 2009: International Symposium on Purinergic Signalling in New Strategy of Drug Discovery; a satellite symposium for the 36th Congress of the International Union of Physiological

Sciences, (2009.7 Fukuoka)

10) Kohro Y, Hasegawa S, Tsuda M, Inoue K. Mechanisms underlying P2X receptor-dependent regulation of neuropathic pain through cPLA2 in DRG neurons: role of PAF receptor and proinflammatory cytokines. Fukuoka Purine 2009: International Symposium on Purinergic Signalling in New Strategy of Drug Discovery; a satellite symposium for the 36th Congress of the International Union of Physiological Sciences, (2009.7 Fukuoka)

11) Shiratori M, Yoshitake M, Tozaki-Saitoh H, Tsuda M, Inoue K, Mechanisms of ATP-induced CXCL2 production and release in mouse microglial cell line, BV2. Fukuoka Purine 2009: International Symposium on Purinergic Signalling in New Strategy of Drug Discovery; a satellite symposium for the 36th Congress of the International Union of Physiological Sciences, (2009.7 Fukuoka)

12) Masuda T, Tsuda M, Inoue K, Interferon-gamma receptor signals are required for spinal microglia activation and neuropathic pain after peripheral nerve injury. Fukuoka Purine 2009: International Symposium on Purinergic Signalling in New Strategy of Drug Discovery; a satellite symposium for the 36th Congress of the International Union of Physiological Sciences, (2009.7 Fukuoka)

13) Tozaki-Saitoh H, Tsuda M, Miyata H, Ueda K, Inoue K, P2Y12 receptors in spinal microglia are required for neuropathic pain after peripheral nerve injury. Fukuoka Purine 2009: International Symposium on Purinergic Signalling in New Strategy of Drug Discovery; a satellite symposium for the 36th Congress of the International Union of Physiological Sciences, invited, (2009.7 Fukuoka)

14) Masuda J, Tsuda M, Tozaki-Saitoh H, K Inoue, Spinal microglia mediate PDGF-induced tactile allodynia in rats. Fukuoka Purine 2009: International Symposium on Purinergic Signalling in New Strategy of Drug Discovery; a satellite symposium for the 36th Congress of the International Union of Physiological Sciences, (2009.7 Fukuoka)

15) Imai T, Nakata E, Kawasaki T, Sakuma S, Yamakawa T, Inoue K, P2X4 receptor- mediated anti-allodynic effect by paroxetine- drug discovery of P2X4 receptor antagonist -. Fukuoka Purine 2009: International Symposium on Purinergic Signalling in New Strategy of Drug Discovery; a satellite symposium for the 36th Congress of the International Union of Physiological Sciences, (2009.7 Fukuoka)

16) Shinozaki Y, Sumitomo K, Tsuda M, Koizumi S, Inoue K, Torimitsu K, Localization of P2X4 receptors in lipid raft-like structure of in vitro model of cell membrane. Fukuoka Purine 2009: International Symposium on Purinergic Signalling in New Strategy of Drug Discovery; a satellite symposium for the 36th Congress of the International Union of Physiological Sciences, Invited, (2009.7 Fukuoka)

17) Nishida M, Ogushi M, Suda R, Nakaya M, Inoue K, Kurose H, Down-regulation of angiotensin type 1 receptor by purinergic P2Y2 receptor stimulation through S-nitrosylation of nuclear factor ・B (NF-・B). Fukuoka Purine 2009: International Symposium on Purinergic Signalling in New Strategy of Drug Discovery; a satellite symposium for the 36th Congress of the International Union of Physiological Sciences, (2009.7 Fukuoka)

18) Nishida M, Sato Y, Nakaya M, Inoue K, Inoue R, Mori Y, Kurose H, Formation of P2Y2 receptor-TRPC5-eNOS signal complex defines ATP-stimulated anti-hypertrophic responses in rat neonatal cardiomyocytes. Fukuoka Purine 2009: International Symposium on Purinergic Signalling in New Strategy of Drug Discovery; a satellite symposium for the 36th Congress of the International Union of Physiological Sciences, (2009.7 Fukuoka)

19) 井上和秀、津田誠, 脊髄ミクログリア活性化と神経因性疼痛. 第３１回日本疼痛学会、(2009.7名古屋)

20) Inoue K, Pain signalling through purinergic receptors of microglia. The 36th Congress of the International Union of Physiological Sciences, (2009.7-8 Kyoto)

21) Tsuda M, Tozaki-Saitoh H, Inoue K, Microglial purinoceptors in the spinal cord and pathological pain. The 36th Congress of the International Union of Physiological Sciences, (2009.7-8 Kyoto)

22) 井上和秀, ミクログリアと神経因性疼痛。第７回整形外科痛みを語る会。
(2009.7福岡)

23) Inoue K, Purinergic signaling in microglia in neuropathic pain. The 22nd Biennial Meeting of the International Society for Neurochemistry (ISN) 2009/APSN 2009, Plenary lecture, (2009.8 Busan,)

24) Inoue K, The function of microglia in neuropathic pain. 2009 European Glial Cell Meeting (Euroglia 2009), (2009.9 Paris)

25) 井上和秀。末梢神経損傷による脊髄ミクログリアの活性化と疼痛。第２５回神経組織の成長・再生・移植研究会、特別講演、大阪(2010.5.22)

26) Kazuhide Inoue. The role of spinal microglia in neuropathic pain. The Lecture to enter as Foreign Academicien member of the Royal Academy of Pharmacy of Spain, Special Lecture, Madrid (2010.5.28)

27) Kazuhide Inoue. The function of microglial P2 purinergic receptors in neuropathic pain. Purine 2010: Adenine Nucleosides and Nucleotides in Biomedicine, invited symposist, Tarragona, Spain (2010, 5.31).

28) Emika Toyomitsu, Makoto Tsuda, Hidetoshi Tozaki-Saitoh, Kazuhide Inoue. CCR2 enhances P2X4 receptor trafficking to the plasma membrane of microglia. Purines 2010 Meeting, poster, Tarragona, Spain (2010.5.30-2010.6.2)

29) Junya Masuda, Hidetoshi Tozaki-Saitoh, Makoto Tsuda, Kazuhide Inoue. Analysis of microglial activation in tissue slice using two-photon microscopy. Purines 2010, Poster-Oral, Spain, Tarragona (2010.5.30-6.2)

30) Ayako Kataoka, Hidetosi Tozaki-Saitoh, Yui Koga, Ayumi Uesugi, Makoto Tsuda, Kazuhide Inoue. Involvement of PKD in UDP-stimulated microglial phagocytosis. Purines 2010 Meeting, poster, Tarragona, Spain (2010.5.30-2010.6.2)

31) Takahiro Masuda, Makoto Tsuda, Ryohei Yoshinaga, Tomohiko Tamura, Kazuhide Inoue. Role of interferon regulatory factor-8 in the pathogenesis of neuropathic pain. Purine2010, oral & poster presentation, Tarragona, Spain (2010.5.30-6.2)

32) 井上和秀. 神経障害性疼痛の基礎：ミクログリアと ATP 受容体研究. 第 32 回日本疼痛学会、シンポジウム、招待、京都 (2010.7.3)

33) 井上和秀、津田誠。神経障害性疼痛発症におけるグリア細胞の役割。Neuro2010 (第 53 回日本神経化学会大会、第 33 回日本神経科学大会、ならびに第20回日本神経回路学会大会の合同大会)、シンポジウム、招待、神戸 (2010.9.2-4)

34) 津田誠，増田隆博，齊藤秀俊，井上和秀。脊髄ミクログリア：難治性慢性疼痛における重要な役者。Neuro2010 (第 33 回日本神経科学大会、第 53 回日本神経化学会大会ならびに第 20 回日本神経回路学会大会)、シンポジウム、招待、神戸 (2010.9.2-4)

35) Kenichiro Nagata, Tomoyuki Inoue, Takayuki Yano, Yuta Kohro, Nobuaki Egashira, Ryozo Oishi, Makoto Tsuda, Kazuhide Inoue. Activation of spinal microglia contributes to paclitaxel-induced mechanical allodynia, cold allodynia and motor dysfunction.

36) 増田隆博、津田誠、吉永良平、田村智彦、井上和秀。Interferon regulatory factor-8 はミクログリア由来疼痛関連分子の発現を誘導する転写因子である。Neuro2010 (第 53 回日本神経化学会大会、第 33 回日本神経科学大会、ならびに第 20 回日本神経回路学会大会の合同大会)、一般、口頭発表、神戸 (2010.9.2-4)

37) 片岡彩子、齊藤秀俊、古賀結衣、上杉歩未、津田誠、井上和秀. UDP によるミクログリアの貪食促進機構における PKD の関与. Neuro2010 (第 53 回日本神経化学会大会、第 33 回日本神経科学大会、ならびに第 20 回日本神経回路学会大会の合同大会)、(ポスター発表)、神戸(2010.9.2-4)

38) 増田潤哉、齊藤秀俊、津田誠、井上和秀。急性脳スライスにおけるミクログリアの状態のリアルタイムイメージング解析。Neuro2010 (第 53 回日本神経化学会大会、第 33 回日本神経科学大会、ならびに第 20 回日本神経回路学会大会の合同大会)、ポスター、神戸 (2010.9.2-4)

39) 高露 雄太, 津田 誠, 矢野 貴之, 辻川 智子, 北野 順子, 齊藤 秀俊, 井上 和秀。神経障害性疼痛の維持機構における STAT3 の役割。Neuro2010 (第 53 回日本神経化学会大会、第 33 回日本神経科学大会、ならびに第 20 回日本神経回路学会大会の合同大会)、ポスター、神戸 (2010.9.2-4)

40) 白鳥美穂、齊藤秀俊、吉武麻衣、津田誠、井上和秀。ミクログリアにおける P2X7 受容体活性化は NFAT と PKC/MAPK を介して CXCL2 産生に関与する。Neuro2010 (第 53 回日本神経化学会大会、第 33 回日本神経科学大会、ならびに第 20 回日本神経回路学会大会の合同大会)、ポスター、神戸 (2010.9.2-4)

41) 山下智大、津田誠、齊藤秀俊、井上和秀。CTP によるミクログリアにおける P2X4 受容体の機能評価。Neuro2010 (第 53 回日本神経化学会大会、第 33 回日本神経科学大会、ならびに第 20 回日本神経回路学会大会の合同大会)、ポスター、神戸 (2010.9.2-4)

42) 下山裕、塚本恵子、齊藤秀俊、津田誠、井上和秀。神経障害性疼痛の緩和における脊髄内 CB2 受容体とその下流メカニズムの重要性。Neuro2010 (第 53 回日本神経化学会大会、第 33 回日本神経科学大会、ならびに第 20 回日本神経回路学会大会の合同大会)、ポスター発表、神戸 (2010.9.2-4)

43) 井上和秀。難治性疼痛の発症メカニズムにおけるグリア細胞の関与。Neuro2010 (第 53 回日本神経化学会大会、第 33 回日本神経科学大会、

ならびに第20回日本神経回路学会大会の合同大会）、ランチョンセミナー、神戸（2010.9.2-4）

44）豊満笑加、津田誠、齊藤秀俊、井上和秀。CCL2シグナルを介したフィブロネクチン刺激によるミクログリア P2X4 受容体の膜移行。Neuro2010（第53回日本神経化学会大会、第33回日本神経科学大会、ならびに第20回日本神経回路学会大会の合同大会）、ポスター、神戸（2010.9.2-4）

45）Kazuhide Inoue. Microglia in pain signalling. The 29th Naito Conference: Glia World, invited, Kanagawa (2010.10.5-8)

46）Takahiro Masuda, Makoto Tsuda, Ryohei Yoshinaga, Tomohiko Tamura, Kazuhide Inoue. Interferon regulatory factor-8 in spinal microglia is a transcription factor crucial for the development of neuropathic pain. Glia World, poster presentation, Kanagawa, Zusi (2010.10.5-8)

47）Emika Toyomitsu, Makoto Tsuda, Hidetoshi Tozaki-Saitoh, Kazuhide Inoue. Upregulation of P2X4R expression on surface of fibronectin-stimulated microglia is mediated by CCL2 signaling. The 29th Naito Conference: Glia World, poster, Kanagawa, Zusi (2010.10.5-8)

48）松村裕太、佐々木淳、津田誠、倉石泰、井上和秀。帯状疱疹痛におけるミクログリアおよびP2X4受容体の役割。第63回日本薬理学会西南部会、口演、鹿児島（2010.11.26）

49）津田誠，井上和秀、痛みの慢性化を抑制するグリア細胞制御。日本麻酔科学会第58回学術集会、神戸、2011.5.19-21

50）Makoto Tsuda, Kazuhide Inoue. Spinal microglia: crucial non-neuronal cells for neuropathic pain. 2nd KPRC Seoul Pain Symposium Program, Seoul, 2011.6.17

51）津田誠，井上和秀。脊髄グリア細胞から探る神経障害性疼痛メカニズム。第16回日本緩和医療学会学術大会、札幌、2011.7.29-30

52）K. Inoue. Glial functions in neuropathic pain. ISN Satellite meeting: Glial cells in (patho) physiology, リュブリャナ（スロヴェニア）Slovenian Academy of Sciences and Arts, 2011/08/23 -- 2011/08/28

53）Makoto Tsuda, Takahiro Masuda, Kazuhide Inoue. Microglia in pain signaling. 10th Euroglia Meeting on Glial Cells in Health and Diseases, Prague, Czech Republic, Sep 13-17, 2011 (Inoue: Organizer)

54）増田隆博，津田誠，吉永遼平，齊藤秀俊，田村智彦，井上和秀。Interferon regulatory factor-8は、神経損傷後に見られる脊髄ミクログリアの過活動状態への移行に重要な転写因子である。第54回日本神経化学会大会、石川県山代、2011.9.26-28

55）吉永遼平，津田誠，増田隆博，西本奈央，齋藤秀俊，田村智彦，井上和秀。脊髄ミクログリアの interferon regulatory factor-5 は神経障害性疼痛の発現に重要な転写因子である。第54回日本神経化学会大会、石川県山代、2011.9.26-28

56）齊藤秀俊，津田誠，井上和秀。初代培養ミクログリアにおける P2Y12 受容体を介したケモカインの発現制御。第54回日本神経化学会大会、石川県山代、2011.9.26-28

57）上杉歩未，片岡彩子，齊藤秀俊，津田誠，井上和秀。UDP 誘発性のミクログリアによるマクロピノサイトーシスへの PKD の関与。第54回日本神経化学会大会、石川県山代、2011.9.26-28

58）K. Inoue, Glia in neuropathic pain after peripheral nerve injury. Milan Pain Symposium "New insights in the pharmacological control of pain"
Milan, 2011/09/29 -- 2011/10/04

59）K. Inoue. Microglial P2 receptor functions in neuropathic pain. The 9th IASP Research Symposium. Shanghai, 2011/10/15 -- 2011/10/18

60）津田誠，井上和秀。神経障害性疼痛とグリア細胞～その役割と分子メカニズム～。第4回日本運動器疼痛学会、大阪、2011.11.19-20

61）増田潤哉、齊藤秀俊、米田聡介、津田 誠、井上和秀。神経障害性疼痛モデルにおいて脊髄ミクログリア特異的に発現する転写因子MafBの役割。第 64 回日本薬理学会西南部会、福岡、2011.11.20

62）津田誠，増田隆博，齊藤秀俊，井上和秀。神経障害性疼痛に重要なミクログリア転写因子。第39回薬物活性シンポジウム、福岡、2011.11.21

63）津田誠，高露雄太，辻川智子，矢野貴之，北野順子，齊藤秀俊，井上和秀．神経障害性疼痛の維持における JAK-STAT3 シグナリングの役割。第39回薬物活性シンポジウム、福岡、2011.11.21

64）米田聡介，増田潤哉，齊藤秀俊，津田 誠，井上和秀．神経障害性疼痛におけるミクログリアの転写因子 MafB の役割。第 39 回薬物活性シンポジウム、福岡、2011.11.21

65）齊藤秀俊，宮田広行，津田 誠，井上和秀。神の関与。第 39 回薬物活性シンポジウム、福岡、2011.11.21

66）Makoto Tsuda, Takahiro Masuda, Kazuhide Inoue. IRF8 is a critical transcription factor required for microglia activation. 第34回分子生物学会、横浜、2011.12.13-16

H．知的財産権の出願・登録状況

なし

Ⅲ．研究成果の刊行に関する一覧表

研究成果の刊行に関する一覧表

書籍

著者氏名	論文タイトル名	書籍全体の編集者名	書　籍　名	出版社名	出版地	出版年	ページ
内田　研造, 馬場　久敏	腰椎椎間孔狭窄に対する顕微鏡下除圧術		OS NOW Instruction 脊椎の低侵襲手術 患者負担を軽減する手術のコツ	株式会社メジカルビュー社		2009	161-171
内田研造, 馬場久敏	外側型腰椎椎間板ヘルニア		アトラス骨・関節画像診断 5.脊椎・脊髄	中外医学社		2011	24-25
内田研造, 中嶋秀明, 馬場久敏	変性疾患；椎間孔狭窄に対する顕微鏡下除圧術		OS NOW Instruction 腰椎の手術 ベーシックからアドバンストまで必須テクニック	株式会社メジカルビュー社		2011	52-57
内田研造, 渡邉修司, 馬場久敏	腰椎		整形外科臨床パサージュ 8 運動器のペインマネジメント	株式会社中山書店		2011	79-87
上野雄文	VBM	加藤敏・神庭重信 他5名	現代精神医学事典	株式会社弘文堂	東京都	2011	86
上野雄文	SPM	加藤敏・神庭重信 他5名	現代精神医学事典	株式会社弘文堂	東京都	2011	106
上野雄文	fMRI (機能的MRI)	加藤敏・神庭重信 他5名	現代精神医学事典	株式会社弘文堂	東京都	2011	112
上野雄文	人工知能	加藤敏・神庭重信 他5名	現代精神医学事典	株式会社弘文堂	東京都	2011	525
上野雄文	ペリオドグラム	加藤敏・神庭重信 他5名	現代精神医学事典	株式会社弘文堂	東京都	2011	948

雑誌

発表者氏名	論文タイトル名	発表誌名	巻号	ページ	出版年
Uchida K, Nakajima H, Yayama T, Kobayashi S, Shimada S, Tsuchida T, Okazawa H, Mwaka E, Baba H.	High-resolution magnetic resonance imaging and 18FDG-PET findings of the cervical spinal cord before and after decompressive surgery in patients with compressive myelopathy	Spine	34(11)	1185-91	2009
Inukai T, Uchida K, Nakajima H, Yayama T, Kobayashi S, Mwaka ES, Guerrero AR, Baba H.	Tumor necrosis factor-alpha and its receptors contribute to apoptosis of oligodendrocytes in the spinal cord of spinal hyperostotic mouse (twy/twy) sustaining chronic mechanical compression	Spine	34(26)	2848-57	2009
Uchida K, Nakajima H, Miyazaki T, Yayama T,Kawahara H, Kobayashi S, Tsuchida T, Okazawa H, Fujibayashi Y, Baba H.	Effects of alendronate on bone metabolism in glucocorticoid-induced osteoporosis measured by 18F-fluoride PET: A prospective study	J Nucl Med	50(11)	11	2009
Uchida K, Nakajima H, Sato R, Yayama T, Erisa Mwaka, Kobayashi S, Baba H.	Cervical spondylotic myelopathy associated with kyphosis or sagittal sigmoid alignment: outcome after anterior or posterior decompression	J Neurosurg Spine	11	5	2009
Kondo T, et.al.	Transient receptor potential A1 mediates gastric distention-induced visceral pain in rats	Gut	58	1342-52	2009
Ushida T, Fukumoto M, Binti C, Ikemoto T, Taniguchi S, Ikeuchi M, et al.	Alterations of contralateral thalamic perfusion in neuropathic pain.	Open Neuroimag	J.4	182-6.	2010

Arai YC, Matsubara T, Shimo K, Suetomi K, Nishihara M, Ushida T, et al.	Low-dose gabapentin as useful adjuvant to opioids for neuropathic cancer pain when combined with low-dose imipramine.	J Anesth	Jun:24(3)	407-10	2010
牛田享宏，井上真輔，大道裕介，神谷光広	【頸部軸性疼痛の病態と治療】 椎体椎間板や前方要素と軸性疼痛	脊椎脊髄ジャーナル	11:23(11)	1003-9	2010
牛田享宏，池本竜則，下和弘，新井健一，西原真理	【神経因性疼痛】 神経障害性疼痛の痛覚認知機構	臨床脳波	10:52(10)	556-63	2010
谷口慎一郎，谷俊一，牛田享宏，永野靖典，田所伸朗	頸椎前方除圧固定術の固定隣接椎間障害による神経障害とX線学的変化の検討.	中部日本整形外科災害外科学会雑誌	09：53(5)	1035-6	2010
谷口慎一郎，谷俊一，牛田享宏，永野靖典，田所伸朗	頸椎後縦靭帯骨化症に対する前方除圧術の周術期合併症	中部日本整形外科災害外科学会雑誌	01：53(1)	59-60	2010
谷口慎一郎，谷俊一，田所伸朗，石田健司，永野靖典，牛田享宏	臨床神経生理学とリハビリテーション 誘発電位による圧迫性頸髄症の機能評価	The Japanese Journal of Rehabilitation Medicine	12:47(12)	842-8	2010
辻貞俊，牛田享宏，新井健一，末冨勝敏，西原真理，池本竜則，et al.	日本神経治療学会 標準的神経治療 慢性疼痛	神経治療学	07：27(4)	591-622	2010
櫻井博紀，牛田享宏	脳波・筋電図の臨床 脊髄・一次求心性線維の機能変化と痛み	臨床脳波	07：52(7)	385-91	2010
Nakajima H, Uchida K, Yayama T, Kobayashi S, Guerrero AR, Furukawa S, Baba H.	Targeted retrograde gene delivery of brain-derived neurotrophic factor suppresses apoptosis of neurons and oligodendroglia after spinal cord injury in rats	Spine	35(5)	497-504	2010

田口敏彦、守屋淳詞	痛みを知る　痛みの疫学	Practice of Pain Management	1(1)	14-20	2010
田口敏彦	腰痛に対するブロック療法	クリニシアン	57(9)	947-952	2010
植松弘進、**柴田政彦**、松村陽子、松田陽一、阪上学、井上隆弥、真下節	当院における脊髄障害性疼痛症例の検討	PAIN RESEARCH	25	19-25	2010
Okubo M, et. al.	Leukotriene synthases and the receptors induced by peripheral nerve injury in the spinal cord contribute to the generation of neuropathic pain	GLIA	58	599-610	2010
Fukuoka T, et. al	Laminae-specific distribution of alpha-subunits of voltage-gated sodium channels in the adult rat spinal cord	Neuroscience	169	994-1006	2010
Okubo M, et. al.	Expression of leukotriene receptors in the rat dorsal root ganglion and the effects on pain behavior	Mol Pain	6	57	2010
Ikemoto T, Kawasaki M, Kato T, Takemasa R, Ushida T, Tani T, et al.	Dangerous cervical radiculopathy by Lemierre's syndrome	J Orthop Sci	4		2011
Ikeuchi M, Ushida T, Izumi M, Tani T.	Percutaneous radiofrequency treatment for refractory anteromedial pain of osteoarthritic knees	Pain Med	12(4)	546-51	2011
Nakamura M, Nishiwaki Y, Ushida T, Toyama Y.	Prevalence and characteristics of chronic musculoskeletal pain in Japan	J Orthop Sci	16(4)	424-32	2011
Nishihara M, Inui K, Motomura E, Otsuru N, Ushida T, Kakigi R.	Auditory N1 as a change-related automatic response	Neurosci Res	71(2)	145-8	2011

Shimo K, Ueno T, Younger J, Nishihara M, Inoue S, Ikemoto T, et al.	Visualization of painful experiences believed to trigger the activation of affective and emotional brain regions in subjects with low back pain	PLoS One	6(11)	e26681	2011
Ushida T, Nishihara M, Arai K, Inoue S.	Conservative therapy for neuropathic pain	Rinsho Shinkeigaku	51(11)	939	2011
Chen KB, Uchida K, Nakajima H, Yayama T, Hirai T, Watanabe S, Guerrero AR, Kobayashi S, Ma WY, Liu SY, Baba H.	Tumor necrosis factor-α antagonist reduces apoptosis of neurons and oligodendroglia in rat spinal cord injury	Spine	36(1)	1350-1358	2011
Chen KB, Uchida K, Nakajima H, Yayama T, Hirai T, Rodriguez Guerrero A, Kobayashi S, Ma WY, Liu SY, Zhu P, Baba H.	High-mobility group box-1 and its receptors contribute to proinflammatory response in the acute phase of spinal cord injury in rats	Spine	36(2)	2122-2129	2011
Sugimoto N, Miwa S, Ohno-Shosaku T, Tsuchiya H, Hitomi Y, Nakamura H, Tomita K, Yachie A, Koizumi.	Activation of tumor suppressor protein PTEN and induction of apoptosis are involved in cAMP-mediated inhibition of cell number in B92 glial cells	Neurosci Lett.	497(1)	55-59	2011
Hirota R, Ngatu NR, Miyamura M, Nakamura H, Suganuma N.	Goishi tea consumption inhibits airway hyperresponsiveness in BALB/c mice	BMC Immunol.	12	45	2011
Usui C, Hatta K, Doi N, Kubo S, Kamigaichi R, Nakanishi A, Nakamura H, Hattori N, Arai H.	Improvements in both psychosis and motor signs in Parkinson's disease, and changes in regional cerebral blood flow after electroconvulsive therapy	ProgNeuropsychopharmacolBiol Psychiatry.	35(7)	1704-1708	2011
Fukutomi Y, Taniguchi M, Watanabe J, Nakamura H, Komase Y, Ohta K, Akasawa A, Nakagawa T, Miyamoto T, Akiyama K.	Time trend in the prevalence of adult asthma in Japan: Findings from population-based surveys in Fujieda City in 1985, 1999, and 2006	Allergol Int.	60(4)	443-448	2011
芦澤健、芦原睦、田口敏彦	痛みと心	Practice of Pain Management	2(3)	148-159	2011

田口敏彦	痛み、しびれの治療　神経ブロック療法	脊椎脊髄ジャーナル	24(5)	403-410	2011
吉田紘二、加藤圭彦、田口敏彦	ラット頸神経の解剖学的検討	整形外科と災害外科	60(1)	13-15	2011
M. Tsuda, Y. Kohro, T. Yano, T. Tsujikawa, J. Kitano, H. Tozaki-Saitoh, S. Koyanagi, S. Ohdo, R-R Ji, M. W. Salter, K. Inoue	JAK-STAT3 pathway regulates spinal astrocyte proliferation and neuropathic pain maintenance in rats	Brain	134(4)	1127-1139	2011
Biber K, Tsuda M, Saitoh-Tozaki H, Tsukamoto K, Toyomitsu E, Masuda T, Boddeke H, Inoue K	Neuronal CCL21 up-regulates microglia P2X4 expression and initiates neuropathic pain development	EMBO J	30	1864-1873	2011
M. Nishida, M. Ogushi, R. Suda, M. Toyotaka, S. Saiki, N. Kitajima, M. Nakaya, K.-M. Kim, T. Ide, Y. Sato, K. Inoue, and H. Kurose	Heterologous down-regulation of angiotensin type 1 receptors by purinergic P2Y2 receptor stimulation through S-nitrosylation of NF-κB	PNAS	108(16)	6662-6667	2011
K. Kuboyama, H. Harada, H. Tozaki-Saitoh, M. Tsuda, K. Ushijima and K. Inoue	Astrocytic P2Y1 receptor is involved in the regulation of cytokine/chemokine transcription and cerebral damage ina rat model of cerebral ischemia	J Cereb Blood Flow Metab.	31(9)	1930-1941	2011
Kataoka A, Koga Y, Uesugi A, Tozaki-Saitoh H, Tsuda M, Inoue K	Involvement of vasodilator-stimulated phosphoprotein in UDP-induced microglial actin aggregation via PKC- and Rho-dependent pathways	Purinergic Signal			2011
K. Kuboyama, M. Tsuda, M. Tsutsui, Y. Toyohara, H. Tozaki-Saitoh, H. Shimokawa, N. Yanagihara and K. Inoue	Reduced spinal microglial activation and neuropathic pain after nerve injury in mice lacking all three nitric oxide synthases	Mol Pain	7	50	2011

Y. Hayashi, K. Kawaji, L. Sun, X. Zhang, K. Koyano, T. Yokoyama, S. Kohsaka, K. Inoue, and H. Nakanishi	Microglial Ca2+-activated K+ Channels Are Possible Molecular Targets for the Analgesic Effects of S-ketamine on Neuropathic Pain	J. Neurosci	31(48)	17370-17382	2011
Tozaki-Saitoh H, Tsuda M, Inoue K	Role of purinergic receptors in CNS function and neuroprotection	Adv Pharmacol	61	495-528	2011
Tsuda M, Tozaki-Saitoh H, Inoue K	Platelet-activating factor and pain	Biol Pharm Bull	34(8)	1159-1162	2011
Inoue K, Tsuda M	Purinergic systems, neuropathic pain and the role of microglia	Exp Neurol			2011
Tsuda M, Tozaki-Saitoh H, Inoue K	Purinergic system, microglia and neuropathic pain	Curr Opin Pharmacol			2011
Aya Nakae, Kunihiro Nakai, Kenji Yano, Ko Hosokawa, Masahiko Shimoda	The animal model of spinal cord injury as an experimental pain model.	Journal of Biomedicine and Biotechnology		11	2011
Nishigami T, Ikeuchi M, Okanoue Y, Wakamatsu S, Matsuya A, Ishida K, et al.	A pilot feasibility study for immediate relief of referred knee pain by hip traction in hip osteoarthritis	J Orthop Sci	23		2012
Uchida K, Yayama T, Sugita D, Nakajima H, Rodriguez Guerrero A, Watanabe S, Roberts S, Johnson WE, Baba H	Initiation and progression of ossification of the posterior longitudinal ligament of the cervical spine in the hereditary spinal hyperostotic mouse (twy/twy)	Eur Spine J	21(1)	149-155	2012
Uchida K, Yayama T, Cai HX, Nakajima H, Sugita D, Guerrero AR, Kobayashi S, Yoshida A, Chen KB, Baba H.	Ossification process involving the human thoracic ligamentum flavum: role of transcription factors	Arthritis Res Ther	13(9)	R144	2012

Uchida K, Nakajima H, Watanabe S, Yayama T, Guerrero AR, Inukai T, Hirai T, Sugita D, Johnson WE, Baba H.	Apoptosis of neurons and oligodendrocytes in the spinal cord of spinal hyperostotic mouse (twy/twy): possible pathomechanism of human cervical compressive myelopathy	Eur Spine J	21(3)	490-497	2012
Cai CH, Yayama T, Uchida K, Nakajima H, Sugita D, Guerrero AR, Yoshida A, Baba H.	Cyclic Tensile Strain Facilitates the Ossification of Ligamentum Flavum Through β-Catenin Signaling Pathway. In Vitro Analysis	Spine	37(11)	639-646	2012
Nakajima H, Uchida K, Guerrero AR, Watanabe S, Sugita D, Takeura N, Yoshida A, Long G, Wright KT, Johnson WE, Baba H.	Blockade of interleukin-6 signaling inhibits the classic pathway and promotes an alternative pathway of macrophage activation after spinal cord injury in mice	J Neuroinflammation		in press	2012
Tadashi Konoshita, Yasukazu Makino, Tomoko Kimura, Miki Fujii, NorihiroMorikawa, Shigeyuki Wakahara, Kenichiro Arakawa, Isao Inoki, Hiroyuki Nakamura, Isamu Miyamori and The Genomic Disease Outcome Consortium (G-DOC) Study Investigators	A crossover comparison of urinary albumin excretion as a new surrogate marker for cardiovascular disease among 4 typesof calcium channel blockers	Int J Cardiol		in press	2012
Tanaka T, Hitomi Y, Kambayashi Y, Hibino Y, Fukutomi Y, Shibata S, SugimotoS, Hatta K, Eboshida A, Konoshita T, Nakamura H.	The differences in the involvements of loci of promoter region andIle50Valin interleukin-4 receptor α chain gene between atopic dermatitis and Japanese cedar pollinosis	Allergol Int		in press	2012
Usui C, Hatta K, Aratani S, Yagishita N, Nishioka K, Kanazawa T, Ito K, Yamano Y, Nakamura H, Nakajima T, Nishioka K.: tom Scale:	The Japanese version of the 2010 American College of Rheumatology Preliminary Diagnostic Criteria for Fibromyalgia and the Fibromyalgia Sympreliability and validity	Mod Rheumatol		in press	2012
竹下克志	運動器疾患における神経障害性疼痛	分子リウマチ治療	5	5-7	2012

Toyomitsu E, Tsuda M, Yamashita T, Tozaki-Saitoh H, Tanaka Y, Inoue K	CCL2 promotes P2X4 receptor trafficking to the cell surface of microglia	Purinergic Signal			2012
井上和秀	痛み研究の最前線	臨床と研究		in press	2012
今井利安、中田恵理子、井上和秀	ギラン・バレー症候群モデル動物における脊髄ミクログリアとP2X4受容体の神経因性疼痛への関与	Pain Research		in press	2012
H. Tozaki-Saitoh, M. Tsuda, K. Inoue	P2Y receptors in microglia and neuroinflammation	Wiley Interdisciplinary Reviews on Membrane Transport and Signaling		in press	2012
井上和秀	神経障害性疼痛とミクログリアのATP活性化イオンチャネル	細胞		in press	2012
井上和秀	神経障害性疼痛におけるCC-Chemokine Ligand 21 (CCL21) の役割	実験医学		in press	2012
井上和秀	痛みとATP	脳21		in press	2012
Okubo M, et. al.	Up-regulation of platelet-activating factor synthases and its receptor in spinal cord contribute to development of neuropathic pain following peripheral nerve injury	Mol Pain		in press	2012
Fukuoka T, et. al.	Re-evaluation of the phenotypic changes in L4 DRG neurons following L5 spinal nerve ligation	Pain	153	68-79	2012

Ⅳ. 研究成果の刊行物・別刷

Journal of Neuroinflammation

BioMed Central

Research

Open Access

Intradermal administration of magnesium sulphate and magnesium chloride produces hypesthesia to mechanical but hyperalgesia to heat stimuli in humans

Takahiro Ushida*[1,2,3], Osamu Iwatsu[2], Kazuhiro Shimo[1], Tomoko Tetsunaga[2], Masahiko Ikeuchi[2], Tatsunori Ikemoto[2,3], Young-Chang P Arai[1], Katsutoshi Suetomi[1] and Makoto Nishihara[1]

Address: [1]Multidisciplinary Pain Center, Aichi Medical University, 21 Karimata, Yazako, Nagakute, Aichi 480-1195, Japan, [2]Department of Orthopaedic Surgery, Kochi Medical School, Kohasu, Okoh-cho, Nankoku, Kochi 783-8505, Japan and [3]Nankoku Pain Research Group, Kochi Medical School, Kohasu, Okoh-cho, Nankoku, Kochi 783-8505, Japan

Email: Takahiro Ushida* - ushidat-koc@umin.ac.jp; Osamu Iwatsu - hcb03073@nifty.com; Kazuhiro Shimo - xia@oct.zaq.ne.jp; Tomoko Tetsunaga - tt1201@softbank.ne.jp; Masahiko Ikeuchi - ikeuchim@kochi-u.ac.jp; Tatsunori Ikemoto - tatsunon31@hotmail.com; Young-Chang P Arai - arainon@aichi-med-u.ac.jp; Katsutoshi Suetomi - suetomi-dm@umin.ac.jp; Makoto Nishihara - nisihara@aichi-med-u.ac.jp

* Corresponding author

Published: 28 August 2009

Journal of Neuroinflammation 2009, **6**:25 doi:10.1186/1742-2094-6-25

Received: 29 April 2009
Accepted: 28 August 2009

This article is available from: http://www.jneuroinflammation.com/content/6/1/25

Abstract

Background: Although magnesium ions (Mg^{2+}) are known to display many similar features to other 2+ charged cations, they seem to have quite an important and unique role in biological settings, such as NMDA blocking effect. However, the role of Mg^{2+} in the neural transmission system has not been studied as sufficiently as calcium ions (Ca^{2+}). To clarify the sensory effects of Mg^{2+} in peripheral nervous systems, sensory changes after intradermal injection of Mg^{2+} were studied in humans.

Methods: Magnesium sulphate, magnesium chloride and saline were injected into the skin of the anterior region of forearms in healthy volunteers and injection-induced irritating pain ("irritating pain", for short), tactile sensation, tactile pressure thresholds, pinch-pain changes and intolerable heat pain thresholds of the lesion were monitored.

Results: Flare formation was observed immediately after magnesium sulphate or magnesium chloride injection. We found that intradermal injections of magnesium sulphate and magnesium chloride transiently caused irritating pain, hypesthesia to noxious and innocuous mechanical stimulations, whereas secondary hyperalgesia due to mechanical stimuli was not observed. In contrast to mechanical stimuli, intolerable heat pain-evoking temperature was significantly decreased at the injection site. In addition to these results, spontaneous pain was immediately attenuated by local cooling.

Conclusion: Membrane-stabilizing effect and peripheral NMDA-blocking effect possibly produced magnesium-induced mechanical hypesthesia, and extracellular cation-induced sensitization of TRPV1 channels was thought to be the primary mechanism of magnesium-induced heat hyperalgesia.

Background

Although magnesium ions (Mg^{2+}) are widely distributed throughout the whole organ, the role of Mg^{2+} in the neural transmission system has not been studied as sufficiently as calcium ions (Ca^{2+}). Much research has mentioned that Mg^{2+} shows a similar physiological attitude to Ca^{2+} and it has been reported that both ions have a membrane-stabilizing effect on nerves [1,2]. In addition, Mg^{2+} is known to act as a competitor to Ca^{2+}, in extracellular matrix [3]. However, the specific role of Mg^{2+} in neurophysiological transmission, especially concerning peripheral somatosensory systems, has been insufficiently focused on and not understood enough.

Among the various studies, the noncompetitive antagonistic action of Mg^{2+} on N-methyl-D-aspartate (NMDA) receptor, a glutamate receptor, was the focus of various reports [4]. Although the role of spinally-located NMDA receptors has been the focus of pain-related research before, NMDA receptors are also known to exist in peripheral nervous system [5]. Carlton et al. reported an increased population of peripheral glutamate receptors in injured peripheral nerve tissue [6]. In a previous study, Iwatsu et al. reported that intradermal administration of NMDA receptor antagonists, ketamin hydrochloride and magnesium sulphate, produces hypesthesia to mechanical stimuli in humans [7]. Therefore, Mg^{2+} may alter neuronal activities both centrally and peripherally.

Concerning therapeutic effects, magnesium sulphate is known to improve types of pain in humans and animals [8]. On the other hand, Mikkelsen et al. reported that intravenous injection of Mg^{2+} (magnesium sulphate) produces heat hyperalgesia in humans [9].

Since much previous research has reported contradictory results, it is necessary to organize human investigation to clarify real changes of sensory experiences after administrations of Mg^{2+}. In our healthy volunteer study, subjects were asked to estimate the degree of pain and the effect of the drug was examined. In addition to noxious mechanical stimulation and noxious radiant heat stimulation, we performed an experiment evaluating innocuous mechanical stimulation, such as tactile sensation, in order to investigate the effect of intradermally applied magnesium sulphate (MS) and magnesium chloride (MC).

Methods

Fifteen healthy volunteers (age ranged from 26 to 34 years, mean: 29 years) were enrolled in sensory testing study after intradermal injection of magnesium ions and another 15 healthy volunteers (age ranged from 22 to 43 years, mean: 28 years) were enrolled in the experiment for examining the effect of local cooling in Mg^{2+} ion induced irritating pain study. All protocols were conducted in accordance with the recommendations outlined in the Declarations of Helsinki and were approved by the local Medical Ethical Committee. All subjects agreed to the study protocols and signed an informed consent form prior to the examination.

Sensory testing study after intradermal injection of magnesium ion

Using the double-blind method, each subject (n = 15) received one intradermal injection of 0.5 M MS (0.1 ml, 524 mOSM) into one anterior ulnar site on the forearm and one injection of physiological saline (0.1 ml, 0.9% NaCl, 290 mOSM) at the same site into the other forearm as a control. At least one week after injection of 0.5 M MS, the same subjects received one injection of 0.05 M MS (0.1 ml, 337 mOSM) and saline in the same way. With 0.05 M MC and physiological saline solutions, they were injected with MC (0.1 ml, 385 mOSM) and physiological saline (0.1 ml) under the same procedure at least one week later. Therefore, each subject received one injection of 0.5 M & 0.05 M MS, and 0.05 M MC, and three injections of saline. Following injection, the resulting effects were evaluated after 1, 10, 20, 30, 45 and 60 min. for the MS site, MC site and NaCl site. The following tests were undertaken in a quiet room and room temperature was maintained at 25°C. Skin surface around the injection (test) area was kept at 34°C by servo-controlled thermal controller (Dantec Dynamics, Skovlunde, Denmark).

Injection-induced irritating pain evaluation

The intensity of irritating pain was evaluated by 100 mm visual analogue scale ('VAS') before and after injection of test solutions.

Tactile sensation evaluation

Using a horse hair brush, tactile sensations at the wheal region formed by intradermal injection and the region of unaffected skin were compared. Rating the sense of normal skin on the same arm as 10, the tactile sensation at the wheal region was evaluated using a numeric rating scale ('NRS').

Tactile pressure threshold test

Using the von Frey filaments, the tactile pressure threshold in intradermally injected area was measured. Prior to this experiment, pressure force of each von Frey filament was calibrated.

Pinch-pain evaluation test

Using an arterial clamp, the pain intensity evoked by pinch at the wheal region formed by intradermal injection and unaffected normal proximal skin were compared. Rating the sense of pain of normal skin on the same arm as 10, the pain intensity at the wheal region was evaluated using NRS. In addition, same pinch (noxious mechanical)

stimulations were applied to the skin, located 1 cm apart from wheal region to check existence of secondary hyperalgesia.

Measurement of intractable heat pain evoking temperature

Thermal stimulation was applied by Peltier probe controller (Intercross-200, Intercross Co., Tokyo, Japan). Tip of the probe (2.5 × 2.5 cm) was applied to injection site and temperature of the probe was serially increased from 30 to 50°C (+0.5°C/sec.). Experimental subjects were instructed to push a button when they experienced intolerant heat pain sensation (The threshold temperature that induces intolerable heat pain). After pushing the button, the temperature of the probe was programmed to automatically return quickly to 30°C.

Changes in injection-induced irritating pain after cooling

After intradermal administration of MS (0.5 M), graded cooling stimuli (from 25 to 9°C: -0.5 – -2.1°C/sec) were applied to the injection site by Peltier probe controller (UDH-300, Unique Medical Co., Tokyo, Japan) (Fig. 1) and the intensity of irritating pain at each test temperature was recorded by NRS.

Statistical Analysis

All data are expressed as mean ± S.E.M Significant changes over time were determined with the Friedman's analysis of variance by ranks followed by post hoc pairwise comparisons.

Results

Local observation and injection-induced irritating pain evaluation

All test solutions containing Mg^{2+} produced flare formation around the injection site and irritating pain. The intensity of irritating pain evaluated by VAS showed a significant increase at 1–10 min after the injection of 0.5 M and 0.05 M MS. Furthermore, MC produced irritating pain at 1 min after injection. (Fig. 2A)

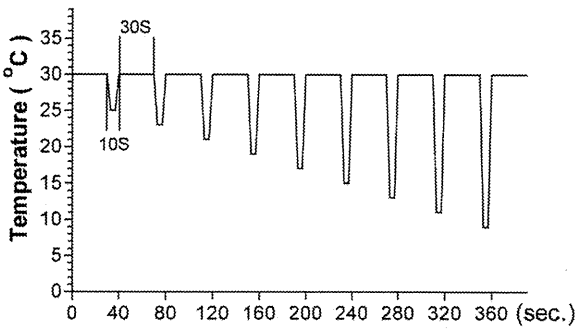

Figure 1
Schematic diagram of the graded cooling stimuli. To check changes in spontaneous pain appearing at the injection site, Peltier probe was directly attached to the injection site and cooled the skin in gradual increments.

Tactile sensation evaluation

After injection of MS, tactile sensation caused by brush decreased up to 20 min and by 1–10 min after injection of 0.5 M and 0.05 M MS respectively, and by 1–10 min after injection of 0.05 M MC (Fig. 2B) compared with the control level.

Tactile pressure threshold test

The tactile pressure threshold measured using the von Frey filaments significantly increased up to 20 min and by 1–10 min after injection of 0.5 M and 0.05 M MS respectively, and up to 20 min after injection of MC compared with saline. (Fig. 2C)

Pinch-pain evaluation test

The pinch-pain evoked by an arterial clamp was reduced up to 10 min after injection of 0.5 M and 0.05 M MS. Similar but shorter changes were observed after injection of MC. (Fig. 2D)

In addition apparent secondary hyperalgesia was not detected in flare area.

Measurement of intolerable heat pain evoking temperature

The threshold temperature that induces intolerable heat pain was decreased up to 20 min and by 1–10 min after intradermal injection of 0.5 M and 0.05 M MS respectively. Similarly MC decreased the pain evoking temperature up to 20 min after injection.

In contrast, intradermal injection of saline did not alter the intolerable heat pain threshold. (Fig. 3)

Effect of local cooling in Mg^{2+} ion-induced irritating pain

Cooling of the surface of the injected area apparently attenuated irritating pain. (Fig. 4) Furthermore, local flare did not change after local cooling.

Discussion

In the present study, both MS and MC solutions but not saline, inhibited the sensations evoked by noxious and innocuous mechanical stimuli but produced irritating pain. Several possible mechanisms can explain the Mg^{2+} ion-induced mechanical hypesthesia. As a general effect of the excitable membrane, changes in external divalent cations are considered to alter membrane surface potential and therefore, a high concentration of Mg^{2+} may inhibit the excitability of the axon by raising the electrical threshold of the membrane directly [2,10]. In pain-related poly-