Purpose
Approximately 20% of the adult population suffer from chronic pain that is inadequately managed with existing analgesics. The lack of effective analgesics stems in part from a lack of appropriate preclinical pain models and from a lack of objective markers that are mechanistically related to pain. The nonhuman primate (NHP) is an ideal preclinical species in that it is neurologically and genetically closer to humans that other preclinical model species. In the current study, pain-related behavior was assessed in NHP models of oxaliplatin-induced neuropathic pain, postoperative pain and in NHP with naturally occurring endometriosis. In addition, brain activity in these pain models was visualized with functional magnetic resonance imaging (fMRI) and analgesics were used to modulate brain activity. Ideally, the current findings would be used not only to further elaborate the mechanism of clinical pain but also to guide the development of clinically useful analgesics.

Methods
Cynomolgus macaques were utilized in creating oxaliplatin-induced neuropathic pain (OXA) and postoperative pain (POP) models. Cynomolgus macaques with endometriosis (EM) were identified based on the presence of abdominal cysts located through ultrasound and confirmation through histopathology. Studies were reviewed and approved by the Hamamatsu Pharma Research, Inc. Animal Care and Use Committee. Baseline responsiveness of the tail to cold water immersion was assessed in awake macaques prior to i.v. infusion of oxaliplatin. The tail was placed in a water bath and the amount of time the tail spent in the water prior to a withdrawal was recorded as the withdrawal latency. In the POP model, under anesthesia, a midline abdominal skin and muscle incision was made. Sensitivity to pressure in awake macaques was assessed using a pressure algometer, both proximally and distally (about 10 cm) to the incision. Similarly, abdominal sensitivity to pressure using a pressure algometer was assessed in macaques with EM.

Under propofol anesthesia, brain activity was visualized using a 3.0T MRI Signa HDxt (GE) with an 8-channel head coil. Brain activity was observed before and during either application of a cold stimulus to the tail (OXA model) or abdominal pressure (POP and EM models). In addition, the effects of analgesic drugs on brain activity during stimulation were observed.

Results

OXA model:
Intravenous infusion of oxaliplatin significantly decreased withdrawal latency (cold hypersensitivity) beginning one day after infusion, lasting for up to three days. Cold hypersensitivity was not observed one week after oxaliplatin infusion. Duloxetine significantly increased withdrawal latency whereas pregabalin and the opioid/monoamine uptake inhibitor tramadol did not. Increased activation of the insular cortex (IC) and the secondary somatosensory cortex (SII) was observed following cold but not warm stimulation. Cold-evoked activation of SII and IC diminished with duloxetine treatment, in line with the antinociceptive effect of duloxetine.

POP model:
Beginning one day following surgery, significant hypersensitivity to pressure emerged both proximally and distally to the incision. Proximal pressure hypersensitivity peaked three days after surgery and responsiveness to pressure was generally at pre-surgery levels one week following surgery. Peak pressure hypersensitivity distal to the incision was observed beginning one day after surgery and persisted for about three days following surgery. Morphine administered one day after surgery alleviated both proximal and distal pressure hypersensitivity in a dose dependent manner. Diclofenac administered two days after surgery did not affect pressure hypersensitivity. Pregabalin administered either one or two days after surgery did not affect pressure hypersensitivity. Significant activation was observed in the anterior cingulate cortex (ACC) and IC one day after surgery. Morphine significantly reduced activation in both ACC and IC. Having no antinociceptive effect, pregabalin significantly reduced ACC activation, but had no effect on IC activation.

EM model:
Pressure response thresholds were significantly decreased in macaques with endometriosis compared to age-matched controls without endometriosis. Morphine increased response pressure whereas acetaminophen and meloxicam did not. Increased activation within the basal ganglia (BG) and IC was observed with pressure applied to the abdomen. Activation in both areas was decreased with morphine. After eight weeks of treatment with dienogest, a progestin clinically used for treatment of endometriosis, pressure hypersensitivity was ameliorated with a general trend in reduction of cyst volume. After eight weeks of treatment, dienogest also decreased pain-related brain activity in BG and IC.

Conclusion
Activation of unique as well as common brain areas appears across NHP pain models and appears to be related to pain. Thus, NHP brain activation could be used as an objective, preclinical marker of pain for various pain states. The current findings also indicate that increased brain activation can be pharmacologically modulated. Interestingly, non-analgesic drugs do not show a similar pattern of inactivation as analgesic drugs, suggesting that brain imaging could also be used as an objective, preclinical marker to guide the development of analgesic drugs and potentially “screen-out” non-analgesic drugs.