CHAPTER V
DISCUSSION AND CONCLUSION

The results have shown that paclitaxel treatment reduced body weight in the P group after the commencement of drug administration compared to the other groups. This is similar to the study of Autheir and coworkers (Autheir et al., 2000) that similar dose regimen of paclitaxel caused significant weight loss since the fourth treatment until the end of the experiment. Interestingly, weight loss as a result of low-dose paclitaxel treatment (5 mg/kg) was also reported (Bardos et al., 2003). Therefore, this weight loss is likely due to the general toxicity of paclitaxel. The mechanism of this toxicity is unknown.

Analgesic test was performed to determine the neuropathy in both tail and hind paw. Using tail flick analysis, significantly prolonged reaction time of tail heat response in the P group was found in week 1 to 3. This finding is similar to another report showing increased reaction time by paclitaxel using the same test (Cavaletti et al., 1997). However, that study also used tail immersion test, another tail heat analgesic test and found no abnormality. This difference may be caused by different techniques used in each experiment. Regarding the hind paw heat analgesic test, significantly increased reaction time in the P group in the third week, similar to the tail heat analgesic test, was seen. Similar result was also observed by Autheir and co-workers (Autheir et al., 2000). As a result, the above evidence indicates that paclitaxel can induce dysfunction of small nerve fibers, especially in the third week after the start of administration. However, in the last two weeks of the experiment, both sensory tests did not show any significant abnormality in the P group relative to the other groups. This may be explained by the recovery of small fibers in that period.

In the electrophysiological test, tail NCV of the P group compared to that of the C group was significantly reduced at week 2 and 5. In the previous

studies, Autheir et al., 2000 and Persohn et al., 2005 have also found the reduction in NCV. Decreased nerve conduction velocity indicates the abnormality of large myelinated fibers, which has been confirmed by histopathological evaluation (Autheir, et al. 2000) and morphometric analysis (Persohn, et al. 2005). Thus, paclitaxel induced reduction in NCV observed in this study was similar to the others. Taken together, this study has confirmed the neuropathy in the rats receiving paclitaxel determined by the analgesic test and NCV study. Accordingly, tissues from these animals can be used to study the MAPK activity.

Gender may have an influence on the susceptibility of rats to develop paclitaxel-associated neuropathy. This study and the study by Authier and co-worker (Authier et al., 2000) have used male rats and observed the similar course of neuropathy after 5 injections of 16 mg/ kg/ week of paclitaxel. In contrast, Chentanez and colleagues (Chentanez et al., 2003) have found similar increase in reaction time from the tail flick test and reduced tail NCV in female rats with lower dose of paclitaxel (5 x 9 mg/ kg/ week). No indicators of neuropathy were seen in the pilot trial using 5 x 9 mg/ kg/ week regimen with male rats which was done prior to this study (data not shown). These data may suggest that female rats are more vulnerable to paclitaxel to develop peripheral neuropathy in comparison with male rats. This hypothesis remains to be confirmed by future studies.

Although the change was statistically insignificant, results from Western blot analysis showed that ERK and p38 phosphorylation was increased in the P group relative to the other groups. This may be because there was no activation of MAPKs in this condition. Another explanation is there was MAPK activation; however, the activation period may occur in early weeks and disappeared at the time when DRGs were removed. To prove this hypothesis, future study of MAPK activity in earlier time points should be conducted. In addition, statistically insignificant changes may be caused by small number of

rats in each group. In this case, increasing number of rats in the future study should also be done.

It is worth nothing that previous studies have reported the neuropathy in the animals induced by the vehicle cremophor (Authier et al., 2000; Gelderblom et al., 2001). However, recent study has found that exposure to paclitaxel but not cremophor is associated with neuropathy in cancer patients (Mielke et al., 2005). In this study, abnormalities in tail flick tests in week 3 and nerve conduction study in week 2 and 5 were observed in the V group compared to the C group, and phosphorylation of p38 in DRG from the V group was elevated to the similar level as seen in the P group. Therefore, the vehicle may partly contribute to the development of neuropathy seen in the P group. However, since the sensory and NCV abnormalities in the P group were more severe than V group, paclitaxel is likely to have additional adverse effects on the peripheral nerve. While the role of cremophor vehicle in neuropathy is still controversial, the results from future studies with paclitaxel dissolved in cremophor should be analyzed with cautions and the vehicle treatment group should be included in the studies.

In conclusion, this study has investigated the phosphorylation of MAPKs which represents their activities in DRG of rats exhibiting neuropathy induced by taxol as indicated by prolonged reaction time when tail and hind paw were tested for heat sensitivity and reduced tail NCV. These findings in paclitaxel-induced neuropathy are similar to those reported by previous studies. Interestingly, some abnormalities in these sensory and nerve conduction tests in the vehicle group were also observed. Westernblot analysis reveals a trend toward increased phosphorylation of ERK in the P group and in both V and P groups for p38. The incompetence to show the significant activation of MAPKs may be due to late removal of DRG for the study of MAPK activity. Further study should be done in earlier time points before the precise activity of MAPKs in sensory neurons in response to paclitaxel can be concluded.

Furthermore, the role of the vehicle, cremophor, and gender in the development of paclitaxel-induced neuropathy should be clarified.