

Poster presentation

Presentation of the work:

The main results of the project work are to be presented on a poster. Each group shall make a poster with the size 0.7 x 1 meter. The posters are to be presented in the corridor between the building 5 and the Hall (outside the auditorium K5).

Monday 22nd November at 1300-1500

At least one of the group members must be in the vicinity of their poster at any time to answer any questions related to the work.

Some practical information:

A short description on how to make a poster is attached. We have no access to a printer that is able to produce paper size 0.7 x 1.0 m format. Therefore you have to print out on A4 or A3 formats and then mount the sheets on a cardboard. You will get a cardboard from the department office. Since you have no access to a color printer, ask your advisor to print out color sheets if needed. Here, the poster should be in portrait format, i.e. height 1m and width 0.7m. You should use portrait format, i.e. height is 1.0 m.

How to make a poster

On engineering and scientific conferences it is very common to use posters to communicate results. Compared to an oral presentation, a poster presentation may be an equally effective way of communicating. Since you are present at your stand, you will meet with the people that have interests in your work. A poster should catch the interest of these people in 3-4 seconds; otherwise they will pass your stand. The design of the poster is therefore very important. Even more important is the content, which should give the impression you have made an investigation of the problem on a relatively high level, and that the results are significant.

The poster will normally contain the following elements:

- Title
- Authors (with affiliation)
- Introduction
- Problem definition and confinement
- Method of investigation
- Results
- Conclusions and recommendations

These items are not necessarily headlines, but rather elements that should be there. How much space you devote to each element will depend on the work that you are presenting.

Some hints:

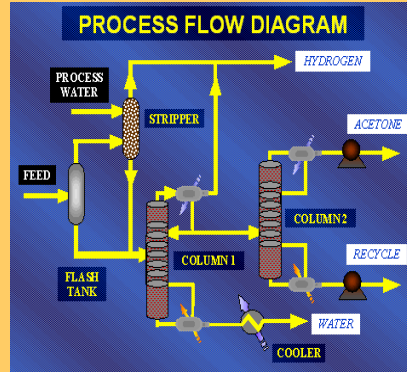
- *Present the problem and results straight forward and simple.* There is not enough space for all details on a single poster. (However, you may bring with you a paper with extra details for those who are interested)
- *The text should be readable from a distance of 2-3 meters.* You should choose a font size and type that is easy to read. The font size of the main text should be minimum 25p, headlines 45-50p and the title about 100p.
- *You may use colors, but do not exaggerate.* Text and colors should have a good contrast to the background. Use complementary colors.
- *Figures, plots and process flow diagrams are important.* Axes must be labeled and have units. Try to simplify without losing too much essential information, in particular process flow diagram (PFD) of large processes. The font size on figures should be on same size as the text.
- *The poster should attract the audience to your stand.* But remember that the conversation with the audience is even more important than the design of the poster.

Attached are two examples of how the poster layout might look like, and two scientific posters.

GOOD LUCK!

Title & Authors

Abstract



Title & Authors

Problem

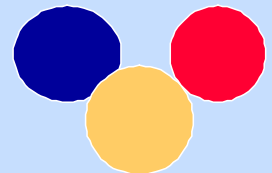
Methods

Results

Table 1

Goals

Fig 2



Conclusions

Activation of p38 kinase in Atlantic salmon macrophages

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The p38 kinase is a member of the Mitogen-activated protein kinase (MAPK) family of proteins. The MAPKs are highly conserved during evolution and are found in all eukaryotes from yeast to mammals [reviewed in 1]. The p38 signalling transduction pathway plays an essential role in regulating many cellular processes including inflammation, cell differentiation, cell growth and death. Activation of p38, often through extracellular stimuli such as bacterial pathogens and cytokines, mediates signal transduction into the nucleus to turn on responsive genes (Fig. 1). The p38 signalling pathway is shown to play an essential role in the production of inflammatory cytokines such as IL-1 β , TNF- α and IL-6 [reviewed in 2].

AIM

To know whether p38 is activated in Atlantic salmon macrophages, we used Western blot analysis with antibodies against the Thr180/Tyr182-phosphorylated form of p38 to detect its activity.

LPS, PMA, synthetic CpG oligodeoxynucleotides (ODNs) and synthetic double-stranded (ds) RNA poly (IC) were tested for their ability to activate p38 in salmon macrophages.

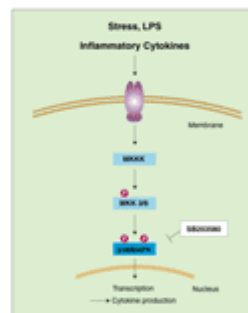


Figure 1: Signals from the cell surface are transduced through the cytoplasm by a cascade of protein kinases: MAPK kinase kinase (MKKK), MAPK kinase 3/6 (MKK3/6) and p38 MAPK. SB 203580 is a selective inhibitor of p38 kinase.

MATERIALS AND METHODS

Materials

Phosphatidylcholine modified CpG ODNs were purchased from Medprobe. Lipopolysaccharide (LPS), the synthetic ds RNA poly (IC) and phorbol myristate phosphate (PMA) were from Sigma. The inhibitor SB203580 was from Alexis.

Western blotting

Atlantic salmon head kidney macrophages were cultivated for 3 days in L-15 medium with 5% FCS. Then medium was replaced with serumfree L-15 and stimulated as indicated in the figure legend.

After stimulation cells were lysed in SDS sample buffer. The cell lysate was resolved by NuPAGE 10% Bis-Tris gel (Novex), followed by transfer to PVDF-membrane (Millipore). The filter was incubated with phosphospecific p38 MAPK antibody [36] (NEW England Biolabs) and immunodetection was performed as instructed by the manufacturer. The membranes were stripped and reprobed with a rabbit polyclonal ab against actin (Sigma) and detected. The Kodak Digital Science TM Image Station was used for making digital images of the membranes. Kodak Image Analysis Software was

used to analyse the images, to enable p38 level to be expressed relative to actin expression in each sample.

Cell protection test for measurement of antiviral activity

CHSE cells treated with supernatants from stimulated leucocytes, were infected with IPNV [5p] at multiplicity of infection (m.o.i.) at 0.01 and 72 hours post infection cytopathic effects (CPE) were measured in microtitre assay [3]. Results are presented as interferon-like cytokine activity (ILC). One unit is defined as the reciprocal dilution at which 50% protection against virus infection is obtained.

RESULTS

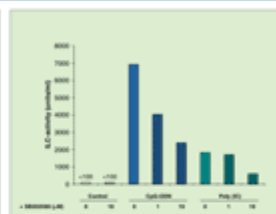
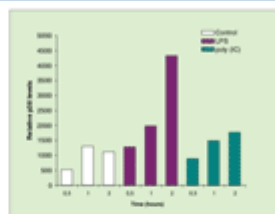
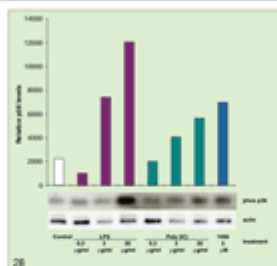
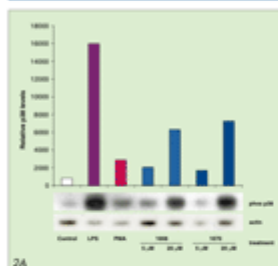


Figure 2A/B: CpG-DNA, LPS, poly (IC) and PMA activate p38-kinase in Atlantic salmon macrophages.

(2A) Atlantic salmon macrophages were stimulated for 2h with CpG ODNs 1668 and 1670 at two concentrations (5 and 20 μ M), LPS (50 μ g ml $^{-1}$) and PMA (200 ng ml $^{-1}$) or

(2B) LPS, poly (IC) and CpG ODN 1668. Controls are untreated cells. After stimulation cells were lysed and subjected to Western blot analysis using antibodies described in M&M. Results shown are representatives of three separate experiments.

Figure 3: Time course of p38 kinase activity in Atlantic salmon macrophages stimulated with LPS or poly (IC).

Macrophages were stimulated with LPS or poly (IC) at 30 μ g ml $^{-1}$ and harvested at indicated time after exposure. After stimulation cells were lysed and subjected to Western blot analysis using antibodies described in M&M.

Figure 4: CpG ODN and poly (IC) induced cytokine release is dependent on p38 activity.

Atlantic salmon leucocytes were preincubated for 1h with the indicated concentrations of the p38 inhibitor SB203580 [4]. Cells with or without SB203580 were then stimulated for 48h with CpG ODN (5 μ M) or poly (IC) (10 μ g ml $^{-1}$). The supernatants were then harvested and assayed for ILC-activity. There was no direct effect of the inhibitor in the concentrations tested on CHSE-cells or the ability of the IPNV virus to replicate in these cells (results not shown).

SUMMARY

- p38 kinase activity can be detected in Atlantic salmon macrophages by a phosphospecific p38 antibody.
- LPS was shown to be the most potent inducer of p38 activity (>15-fold induction), followed by CpG ODNs (>8-fold induction), poly (IC) (>2-fold induction) and PMA.
- p38 activation was detectable 30 min after stimulation, and there was a further increase in activation after 2h of stimulation.
- Preincubation of leucocytes with the specific p38 MAP kinase inhibitor, SB203580, inhibited production of interferon like cytokines by CpG ODNs and poly (IC). This finding suggest that p38 MAPK activation participate in signalling pathways in Atlantic salmon leucocytes.

References

- Williams, C., Gibson, S., Inge, M. B. & G. L. Johnson (1995) Mitogen-Activated Protein Kinase: Conservation of a Three Kinase Module From Yeast to Human. *Physiological Reviews*, 75, 1230-1234.
- Chen, X. & L. Han (2003) The p38 signal transduction pathway: Activation and function. *Cellular Signalling* 15, 1-13.
- Rausch, T., Smith, C. & P. de Kozak (1991) Spectrophotometric method for titration of trout interferon, and its application to routine test by experimentally infected with viral haemorrhagic septicemia virus. *Diseases of Aquatic Organisms*, 10, 27-29.
- Correia, A., Rouse, J., Davis, J. N., Hesse, J., Cohen, J., Collingford, J., Young, P. & C. C. (1995) SB203580 is a specific inhibitor of a MAP kinase homologue which is stimulated by cellular stress and interleukin 1. *FEBS letters*, 364, 229-233.



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