The Role of Prostacyclin (PGI_2) and Thromboxane A_2 (TXA_2) in Pathogenesis of Dengue Hemorrhagic Fever (DHF)

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Abstract

In previous studies it has been demonstrated that the levels of plasma 6 keto-prostaglandin $F_1\alpha$ (6-K-PGF_1), the stable metabolite of PGI_2 were elevated in DHF patients during shock. In this study it is hypothesized that excessive PGI_2 production plays a very important role in developing serious clinical manifestations of dengue shock syndrome (DSS) patients. In addition, an attempt was made to determine whether TXA_2 has any significant role in such patients. Plasma 6-K-PGF_1 and thromboxane B_2 (TXB_2), the stable metabolites of TXA_2 were determined in 43 normal healthy children (NC) and 54 DHF patients without shock (DHF-N) and 33 DHF patients with shock (DHF-S). Subjects aged between 2 and 14 years. Plasma 6-K-PGF_1 and TXB_2 were measured by radioimmunoassay and the ratio of TXB_2/6-K-PGF_1 were also calculated. In 43 NC the values of plasma TXB_2, 6-K-PGF_1 and TXB_2/6-K-PGF_1 ratio were (mean $\pm$ SE) 372.3 $\pm$ 17.1, 150.1 $\pm$ 2.4 and 2.52 $\pm$ 0.12 pg/ml, respectively. In 54 DHF-N patients the corresponding values were 409.1 $\pm$ 16.0, 278.4 $\pm$ 11.6 and 1.54 $\pm$ 0.06 pg/ml; whereas those in 33 DHF-S patients were 254.3 $\pm$ 26.2, 349.1 $\pm$ 20.5 and 0.757 $\pm$ 0.073 pg/ml, respectively. Plasma 6-K-PGF_1 levels of DHF-N and DHF-S patients were significantly greater than those in normal children ($p < 0.001$, $p < 0.01$ respectively). The plasma 6-K-PGF_1 levels seem to be greater in DHF-S patients than in the DHF-N patients, however the difference in values were not statistically significant ($p > 0.05$). These findings indicate that plasma PGI_2 level is significantly increased in DHF particularly during shock. Plasma TXB_2 levels of DHF-N had no significant statistical difference from those of NC ($p > 0.05$); however, those in DHF-S patients were significantly lowered ($p < 0.001$) than those of NC and DHF-N patients. The findings suggest the important role of TXA_2 to compensate for excessive PGI_2 secretion in DHF patients. The failure or inadequate TXA_2 production may eventually lead to shock. The ratios were significantly reduced in both DHF-N and DHF-S patients when compared to those of NC ($p < 0.001$ both). The ratio in DHF-S patients was also significantly lowered than that in DHF-N patients ($p < 0.001$). It is suggested that the imbalance between TXA_2 and PGI_2 production exists during DHF infection. The more reduction of plasma TXA_2/PGI_2 ratio leads to more overt and serious clinical manifestations of the disease.

Key word : Prostacyclin, Thromboxane, Dengue Hemorrhagic Fever, Dengue Shock Syndrome

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Prostacyclin (PGI_2) and thromboxane A_2 (TXA_2) are prostanoids which exert antagonistic actions on vessel walls and blood platelets, and thus play very important roles in vascular homeostasis and blood supply. PGI_2 is the major arachidonic product of vascular endothelium[1,2]; however, it can be secreted by other tissues, i.e. vascular smooth muscle[2], and lymphocytes[3] by interaction with platelets. The biologic actions of prostacyclin include vasodilatation[4-6], hypotension[7], platelet aggregation inhibition[4,6], opposition of leukocytes migration to damaged vascular endothelium[7,8], inflammatory reaction[9-11], increased vascular permeability[11,12], increased organ blood flow and vascular capacity[12,13], and fibrinolytic activity[8]. Clinical manifestations of patients with dengue hemorrhagic fever mimic those of PGI_2 hypersecretion, i.e. fever, extravasation of fluid to the intracellular space leading to organ edema or congestion, hypovolemic hypotension and shock, platelet dysfunction, and hemorrhagic manifestations. In a previous study, it was shown that plasma PGI_2 levels were elevated in dengue hemorrhagic fever with shock[14].

TXA_2 is the major arachidonic acid metabolite of platelets[4]; however, it can be secreted by vascular tissues[5]. This prostanoid exerts opposite actions to prostacyclin on blood vessels and platelets[14], therefore it may play certain roles in dengue shock syndrome. Moncada and Vane[6] have pointed out that an equilibrium between these 2 major prostanoids is an important role in the maintenance of vascular tone and platelet homeostasis.

The purpose of this study is to determine the roles of prostacyclin and thromboxane A_2 in patients with dengue hemorrhagic fever (DHF) and dengue shock syndrome (DSS).

MATERIAL AND METHOD

All subjects of both sexes aging from 2 to 14 years were included in this study. They were divided into 3 groups.

Group I 43 normal children (NC) who were healthy and afebrile.

Group II 54 patients who were admitted to Ramathibodi Hospital with clinical diagnosis of dengue hemorrhagic fever (DHF-N). Their blood samples were obtained during normotension.

Group III 33 dengue hemorrhagic fever patients, whose blood samples were taken during impending shock (DHF-S), i.e. systolic blood pressure was below 90 mmHg and pulse pressure was less than 20 mmHg.

All patients had serology positive for acute dengue virus infection interpreted from two blood samples determinated 2 weeks apart. None of the subjects had received corticosteroids or nonsteroidal antiinflammatory drugs before blood collections.

Methods

Blood samples were collected in tubes containing EDTA and indomethacin. Plasma was separated immediately and stored at –20°C until analysed. Plasma samples were determined for 6-keto prostaglandin $F_1\alpha$ (6-K-PGF_1), the stable metabolite of PGI_2 and thromboxane B_2 (TXB_2), the stable metabolite of TXA_2 by radioimmunoassay method following the same technique we previously used[14].

RESULTS

The mean ± SE of plasma TXB_2 levels of 43 NC, 54 DHF-N, and 33 DHF-S were shown in scattogram (Fig. 1). The values (mean ± SE) were 372.3 ± 17.06, 409.1 ± 16.0 and 254.3 ± 26.2 pg/ml, respectively. The mean plasma TXB_2 levels of 54

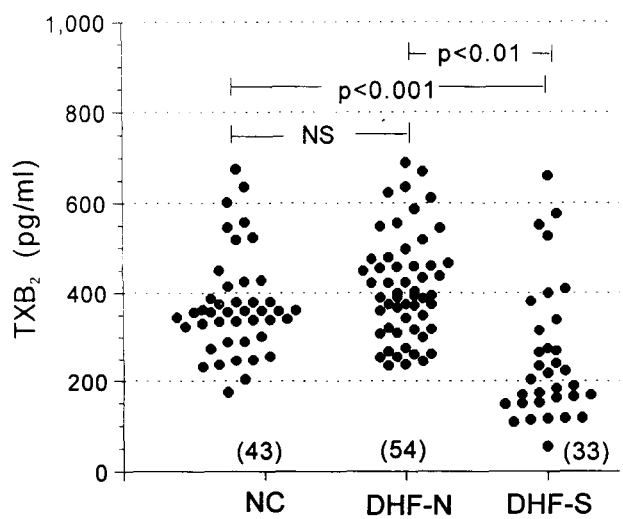

Fig. 1. Plasma thromboxane B2 in normal children and in patients with DHF.

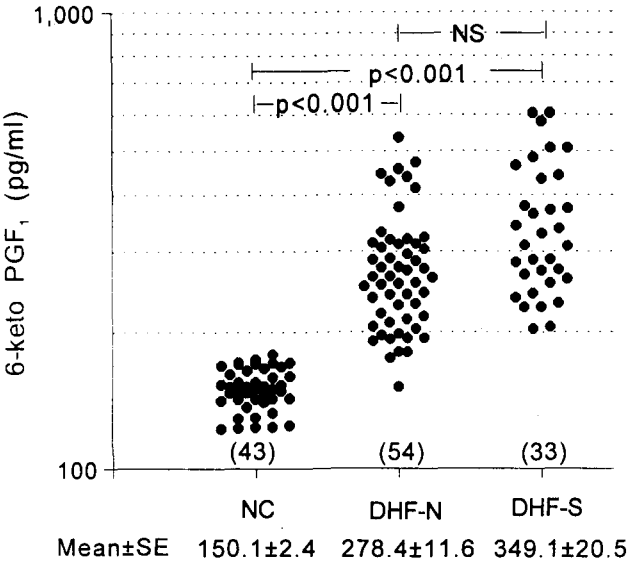

Fig. 2. Plasma 6-keto PGF_1 in normal children and in patients with DHF.

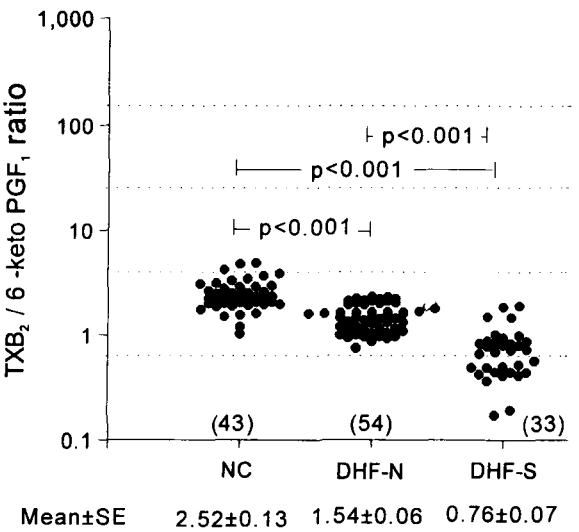

Fig. 3. Plasma TXB_2/6-keto PGF_1 ratio in normal children and in patients with DHF.

DHF-N patients and 43 NC were not statistically different. In contrast, the mean plasma TXB_2 levels of 33 DHF-S patients were significantly lower than those of 43 NC and 54 DHF-N patients ($p<0.001$).

The mean ± SE of plasma 6-K-PGF_1 levels (mean ± SE) of 43 NC, 54 DHF-N and 33 DHF-S (Fig. 2) were 150.1 ± 2.4, 278.4 ± 11.6 and 349.1 ± 20.5 pg/ml, respectively. It was obvious that the plasma 6-K-PGF_1 levels in both groups of dengue hemorrhagic fever patients were statistically higher than those of normal children ($p < 0.001$). The levels were even higher in DHF-S than those of DHF-N; however, they were not significantly different.

The plasma TXB_2/6-K-PGF_1 ratios of 3 groups of subjects were demonstrated in Fig. 3. The ratios of DHF-N and DHF-S patients were significantly lower than those of normal children ($p < 0.001$) and the ratio of DHF-S patients was significantly lower than those of DHF-N patients ($p < 0.001$).

DISCUSSION

It was demonstrated in this study that plasma 6-K-PGF_1 levels were significantly increased in patients with dengue hemorrhagic fever particularly during impending shock. The plasma TXB_2 levels were not changed in dengue hemorrhagic fever without shock; however, the levels were significantly decreased during impending shock. In shock state the plasma levels of 6-K-PGF_1 were definitely increased, whereas the plasma TXB_2 levels were significantly decreased. The findings had pointed out the opposite role of TXA_2 and PGI_2 in pathogenesis of dengue hemorrhagic fever with shock. The ratios of plasma TXB_2 and PGI_2 levels were reduced in dengue hemorrhagic fever patients which were more prominent during state of shock. The findings support the pathogenesis that equilibrium between TXA_2 and PGI_2 is an important role in maintenance of vascular tone and blood pressure regulation.

Now the questions come up that how and why the plasma TXA_2 levels were declined, whereas the plasma PGI_2 levels were elevated in dengue hemorrhagic fever during impending shock. The explanations for this discrepancy are as follow. Firstly, interferon, a cytokine produced by activated lymphocytes, monocytes and neutrophils during acute viral infection has been shown to enhance vascular endothelial PGI_2 synthesis without affecting platelet aggregation and TXA_2 production[15].

This can result in decreased plasma TXA_2/PGI_2 ratio in patients with dengue hemorrhagic fever. Secondly, several mediators released from activated blood cell components such as platelets and leucocytes during dengue virus infection cause leucocyte and macrophage adherence to blood vessel walls and subsequently stimulate the adjacent intact vascular endothelia to synthesize PGI_2[16-24]. Moreover, some of prostaglandin endoperoxides (PGG_2, PGH_2) released from activated platelets and leucocytes are capable of incorporation into PGI_2 by the intact vascular endothelium[25,26]. This may cause further elevation of plasma PGI_2 during dengue virus infection. In contrast, the circulating prostaglandin endoperoxide trapped by the platelets cannot corporate into TXA_2 although it may be corporated into lipoxygenase products[26]. Thirdly, thrombocytopenia during the course of dengue virus infection has limited ability of the platelets to release TXA_2 into systemic circulation. Such events can play a role in decreased plasma TXB_2/6 K-PGF_1 ratio. Fourthly, lymphocytes which contain little or no cyclo-oxygenase activity; but PGI_2 synthase activity can convert prostaglandin endoperoxides which were released from activated platelets and other related cells into PGI_2[3]. This ability of circulating lymphocytes to secrete PGI_2 but not of TXA_2 causes further reduction in plasma TXB_2/6-K-PGF_1 ratios. All of these evidences tend, therefore, to explain the decreased plasma TXB_2/6-K-PGF_1 ratio in dengue hemorrhagic fever patients especially during hypotensive crisis.

In previous study[14], it was hypothesized that excessive PGI_2 production in DHF is the protected mechanism against further endothelial injury[15] and platelet aggregation inhibition by various mediators. Excessive PGI_2 production is expected to prevent thrombosis and enhance thrombolysis. However, excessive PGI_2 production with decreased plasma TXA_2 levels (reduced plasma TXA_2/PGI_2 ratio) can adversely cause generalized vasodilatation, organ congestion and failure, systemic hypotension, profound bleeding, and shock. The extravasation fluid leakage would, therefore, result in dehydration and hemoconcentration. Excessive fluid replacement without correction of decreased plasma TXA_2/PGI_2 ratio is a harzardous procedure leading to progressive extravascular

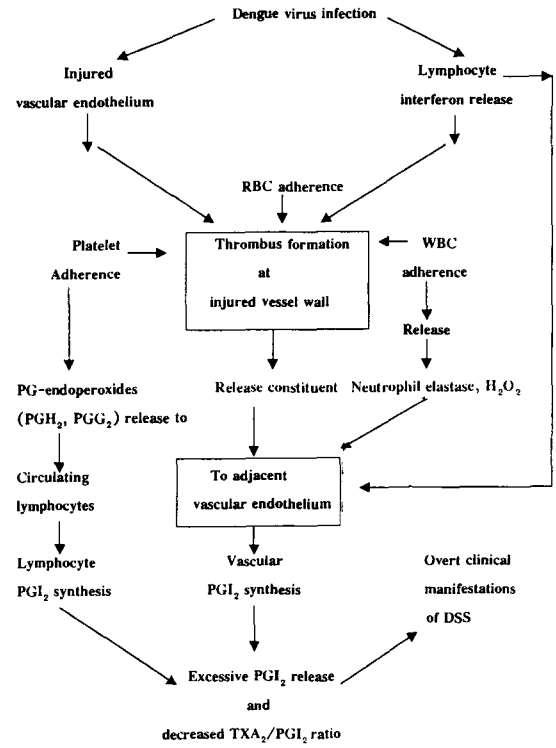

Fig. 4. The role of PGI_2 in the pathogenesis of dengue shock syndrome.

leakage of fluid resulting in organ congestion, edema and failure-particularly the liver, heart, lungs and brain. These events can explain the serious complications such as pericardial, pleural and abdominal effusions, liver and cardiac congestion, as well as pulmonary and brain edema found in terminal state of dengue hemorrhagic fever after huge fluid replacement therapy. The correction of decreased TXA_2/PGI_2 ratios seems to be theoretically more effective than high molecular fluid replacement (such as plasma or dextran) in management of dengue hemorrhagic fever patients during shock state. Fig. 4 is our proposed pathogenesis concerning the role of PGI_2 in dengue shock syndrome.

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บทบาทของ prostacyclin (PGI_2) และ thromboxane A_2 (TXA_2) ต่อการเกิดโรคไข้เลือดออก

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จากการศึกษาครั้งก่อนพบว่าระดับ 6-keto-prostaglandin $F_1\alpha$ (6-K-PGF_1) ซึ่งเป็น metabolite ของ PGI_2 ในเลือดเพิ่มขึ้นในผู้ป่วยไข้เลือดออกที่อยู่ในภาวะช็อก ฉะนั้นจึงบ่งชี้ว่าการสร้าง PGI_2 ในปริมาณมากผิดปกติน่าจะเป็นสาเหตุสำคัญที่ทำให้ผู้ป่วยไข้เลือดออกมีอาการแสดงรุนแรง ในการศึกษาครั้งนี้คณะผู้รายงานต้องการทราบบทบาทของ TXA_2 ในการเกิดโรคนี้ด้วย โดยการวัดระดับ 6-K-PGF_1 และ thromboxane B_2 (TXB_2) ซึ่งเป็น metabolite ของ TXA_2 ในเลือดของเด็กปกติ 43 ราย, เด็กที่ป่วยด้วยโรคไข้เลือดออกที่ไม่มีอาการช็อก (DHF-N) 54 ราย และที่มีอาการช็อก (DHF-S) 33 ราย เด็กทั้ง 3 กลุ่มมีอายุอยู่ระหว่าง 2-14 ปี ผลการศึกษาพบว่าเด็กปกติมีระดับ TXB_2, 6-K-PGF_1 และอัตราส่วนระหว่าง TXB_2/ 6-K-PGF_1 ได้ค่า (ค่าเฉลี่ย $\pm$ ความคลาดเคลื่อนมาตรฐาน) 372 $\pm$ 17.1, 150.1 $\pm$ 2.4 และ 2.52 $\pm$ 0.12 pg/ml ตามลำดับ สำหรับผู้ป่วย DHF-N 54 ราย ได้ค่าดังกล่าว 409.1 $\pm$ 16.0, 278.4 $\pm$ 11.6 และ 1.54 $\pm$ 0.06 pg/ml ในผู้ป่วย DHF-S 33 ราย ได้ค่า 254.3 $\pm$ 26.2, 349.1 $\pm$ 20.5, และ 0.757 $\pm$ 0.073 pg/ml ตามลำดับ ระดับ 6-K-PGF_1 ในเลือดของผู้ป่วย DHF-N และ DHF-S สูงกว่าในเด็กปกติอย่างชัดเจน ($P < 0.001$, $P < 0.01$ ตามลำดับ) ระดับ 6-K-PGF_1 ในผู้ป่วย DHF-S จะสูงกว่าผู้ป่วย DHF-N แต่ยังไม่มีความแตกต่างทางสถิติชัดเจน ($P > 0.05$) ผลการศึกษาบ่งชี้ว่าระดับ PGI_2 เพิ่มขึ้นในเลือดของผู้ป่วยไข้เลือดออกโดยเฉพาะอย่างยิ่งในภาวะช็อก ส่วนระดับ TXB_2 ในผู้ป่วย DHF-N และเด็กปกติไม่แตกต่างกันอย่างมีนัยสำคัญ ($P > 0.05$) แต่ถึงกระนั้นก็ตามระดับ TXB_2 ในผู้ป่วย DHF-S ก็ยังต่ำกว่าในเด็กปกติและ DHF-N ($P < 0.001$) จึงสรุปได้ว่า TXA_2 ที่หลั่งเพิ่มขึ้นในโรคไข้เลือดออกนั้นเพื่อต่อต้านฤทธิ์ของ PGI_2 ไม่ให้รุนแรงเกิน เมื่อใดที่การหลั่งของ TXA_2 ไม่เพียงพอต่อการเพิ่มขึ้นอย่างมหาศาลของ PGI_2 แล้วผู้ป่วยก็จะมีอาการช็อก อัตราส่วนระหว่างระดับ TXB_2/6-K-PGF_1 ในเลือดจะลดลงอย่างชัดเจนในผู้ป่วย DHF-N และลดลงอย่างมากในผู้ป่วย DHF-S เมื่อเทียบกับเด็กปกติ การเสียดุลระหว่างการสร้าง TXA_2 และ PGI_2 ทำให้เกิดอาการแสดงของไข้เลือดออก อาการยิ่งมากขึ้นเมื่ออัตราส่วนดังกล่าวลดลงมาก

คำสำคัญ : โรคไข้เลือดออก, โปรสตาซัยคลิน, ธรอมบอกเซน

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