CHAPTER 6

MATHEMATICAL MODEL OF FOUR SECRETION HORMONES IN HEALTHY MALE SUBJECTS

6.1 Introduction

Due to the complexity of delay differential equations, many scientists did not include them in their models. However, many biological processes have inherent delays and including them may lead to additional insights in the study of complicated biological processes. The neuroendocrine challenge paradigm is based on the involvement of monoamine pathways in the control of anterior pituitary hormone secretion and has been described extensively. Psychotropic drugs with different effects on the central neurotransmitter system have distinct effects on the anterior pituitary hormone secretion and can be characterized by certain pharmaco endocrinological profiles.

Antidepressants which primarily act via noradrenaline (NA)-reuptake inhibition (e.g. desipramine) stimulate gonotropin release hormone (GnRH)-secretion **[20,146],** whereas serotonin (5-HT)-reuptake inhibiting antidepressants (e.g. indalpine, chlomipramine) are characterized by prolactin (PRL)-

stimulation. Cortisol (COR) secretion can acutely be increased by antidepressants with both NA or 5-HT reuptake inhibition; the stimulatory effects of antidepressants on COR secretion are mediated via stimulation of the ACTH output of the pituitary gland **[45].**In this study the COR, ACTH-, GnRH-, and PRL-secretion after oral administration of 4 mg reboxetine was measured and compared to placebo.

Reboxetine is a novel antidepressant drug that selectively inhibits NA reuptake (IC 50 values: NA reuptake8 nM; 5-HT reuptake 1070 nM; functional selectivity 5-HT/NA 130; DA reuptake10 000 nM). Unlike desipramine or imipramine, reboxetine has only weak affinity (K_i > 1000 nmol/l) for muscarinergic, histaminergic H1, adrenergic α_1, and dopaminergic D_2 receptors. Moreover, reboxetine1 2 is rapidly absorbed in man (t_{max} about 3 h) with a terminal elimination half-life of 13 h and has linear pharmacokinetics in young, healthy males for single doses of 1–5 mg. The absolute bioavailability of reboxetine enantionmers is 0.919 and 1.02 for R, R (-) reboxetine and S, S (+) reboxetine, respectively. In clinical studies, reboxetine has been shown to have a favorable tolerability and safety profile with only few side effects such as dry mouth, constipation, increased sweating, insomnia, urinary hesitancy or retention, and tachycardia.

Furthermore, in placebo-controlled and active comparator trials the antidepressant efficacy of reboxetine has been demonstrated. A system of

differential equations serves as the mathematical model, describing the dynamics of hormones. Since the processes take place in different parts of the body and influence each other with a certain delay, passing over to delay differential equations is deemed a reasonable step. A stochastic process is chosen to model the time points of this COR, ACTH, GnRH and PRL generator. Focus in this paper is on the model development. This rather elaborate mathematical model is the basis for a detailed.

Twelve healthy male subjects of normal weight, age 20–35 years, were included upon receipt of their informed consent, following a clinical examination (psychiatric and medical history, physical examination) and establishment of normal laboratory parameters (Hb, K^+, Na^+, glutamate oxalo acetate transaminase (GOT), glutamate pyruvate transaminase (GPT), γ- GT, blood sugar, bilirubin, serum creatinine, heart rate, electrocardiogram and electroencephalogram). Alcohol abstinence 24 hours prior to each experiment and abstinence from medication beginning four weeks before the study were mandatory.

Each subject took part two times in the trial; in a randomized order the volunteers received placebo or reboxetine (4 mg orally) on two different days. At 0700 h, an IV catheter was inserted into the antecubital vein and kept open with physiological saline solution. The subjects rested in bed throughout the experiments (up to t = 390 min). At t = 60 min (0700 h), t = 0 min (0800 h;

administration of placebo or reboxetine), and at intervals of 30 minutes thereafter up to t = 390 min (1300 h), blood was drawn. Subjects fasted from completion of the evening meal the day before until conclusion of the experiment **[45].**

6.2 Mathematical model and Assumptions

In formulating the mathematical model of four secretion hormones, the following events are considered. The interactions among the hormones are all composed of complex enzyme reactions following cooperative kinetics rather than Michaelis Menten type.

In forming the growth equations of hormones COR(C), ACTH(A), GnRH(G) and PRL(P)-secretion, taking into account the effects of hormones**[6].** A system of differential equations serves as the mathematical model, describing the dynamics of hormones.

The rate of growth of the hormones due to corresponding feedbacks are represented by the nonlinear term of the form $\frac{A_1}{K_1 + T^m(t - \Upsilon_1)}$ where X (t) denotes the effects on COR, ACTH, GnRH, and PRL-secretion and m>0 is called the Hill coefficient, which is related to the degree of cooperatively of ligand hormones with the specific binding protein and Υ_i.

Rate equations of growth of the reproductive hormone are written in the

form of following nonlinear delay differential equations:

$$\tilde{C}(t) = -\alpha C(t) + \frac{A_1}{K_1 + T^m(t-\Upsilon_1)},$$

$$\tilde{A}(t) = -\beta A(t) + \frac{A_2}{K_1 + P^m(t-\Upsilon_3)} + A_2 C(t-\Upsilon_2),$$

$$\tilde{P} = -\delta P(t) + A_4 A(t-\Upsilon_4), \qquad 6.1$$

$$\tilde{G}(t) = -\gamma G(t) + A_3 C(t-\Upsilon_3).$$

Where $A_1 A_2 A_3 A_4$ are the respective rates of synthesis of C, A ,G, and P hormones, and $\alpha, \beta, \delta, \gamma$ are the decay rates of C ,A, G,and P in the blood stream. It is assumed that each of these hormones is cleared from the blood stream according to the first order kinetics **[6, 65,127,141].**

$$C(t) = \Omega_1(t), for - \Upsilon_1 \le t \le \infty,$$

$$A(t) = \Omega_2(t), for - \Upsilon_2 \le t \le \infty,$$

$$G(t) = \Omega_3(t), for - \Upsilon_3 \le t \le \infty, \qquad 6.2$$

$$P(t) = \Omega_4(t), for - \Upsilon_4 \le t \le \infty.$$

Where $\Omega_i(t) \in \vec{C}([-\Upsilon_{i,} 0], \infty)$

It is need to study the stability of Reboxetine or Placebo and the solution of the system of equations

Dimensionless System

$$\text{Let} \quad x=\frac{C}{\alpha_1}, x=\frac{F}{\alpha_2}, x=\frac{G}{\alpha_3}, x=\frac{P}{\alpha_4}, x=\frac{t}{\alpha_5} \tag{6.3}$$

(6.1) becomes

$$\dot{x}(t)=-\alpha\alpha_5 x(t)+\frac{\frac{A_1\alpha_5}{\alpha_1{\alpha_3}^m}}{\frac{K_1}{\alpha_3^m}+z^m(t-\Upsilon_1)}$$

$$\dot{y}(t)=-\beta\alpha_5 y(t)+\frac{A_2\alpha_1\alpha_5}{\alpha_2}x(t-\Upsilon_2)+\frac{\frac{A_1\alpha_5}{\alpha_2{\alpha_4}^m}}{\frac{K_1}{\alpha_4^m}+w^m(t-\Upsilon_3)} \tag{6.4}$$

$$\dot{z}(t)=-\gamma\alpha_5 y(t)+\frac{A_4\alpha_1\alpha_5}{\alpha_3}y(t-\Upsilon_3)$$

$$\dot{w}(t)=-\delta\alpha_5 w(t)+\frac{A_5\alpha_2\alpha_5}{\alpha_4}y(t-\Upsilon_4)$$

Where

$$\alpha_1=m\sqrt{K_1}\left(\frac{A_1}{A_3}\right)^2\left(\frac{A_2}{A_5}\right),\ \ \alpha_2=m\sqrt{K_1}\left(\frac{A_2A_1}{A_5A_3}\right),$$

$$\alpha_3=m\sqrt{K_1},\alpha_4=m\sqrt{K_1},\ \ \alpha_4=\frac{A_3}{A_1A_2}$$

The dimensionless equations are

$$\dot{x}(t) = -a_1 x(t) + \frac{A}{1 + z^m(t - \Upsilon_1)},$$

$$\dot{y}(t) = -a_2 y(t) + x(t - \Upsilon_2) + \frac{A}{1 + w^m(t - \Upsilon_3)},$$

$$\dot{z}(t) = -a_3 z(t) + Ky(t - \Upsilon_3),$$

$$\dot{w}(t) = -a_4 w(t) + y(t - \Upsilon_4). \qquad 6.5$$

Where

$$a_1 = \frac{A_3\alpha}{A_1 A_2}, a_2 = \frac{A_3\beta}{A_1 A_2}, a_3 = \frac{A_3\gamma}{A_1 A_2}, a_4 = \frac{A_3\delta}{A_1 A_2},$$

$$A = \frac{A_3^3 A}{A_1^2 A_2^2} \frac{1}{K_1^{(m+1)/m}} \quad and \quad K = \frac{A_5 A_4}{A_3 A_5}$$

6.3 Existence of steady states of the system

Let $\hat{E}(\hat{x}, \hat{y}, \hat{z}, \hat{w})$ be a steady state of the system in Reboxetine or Placebo $\hat{x} = \left(\frac{a_3 \hat{z}}{K}\right), \hat{y} = a_4 \hat{w}$ where $\hat{z}$ and $\hat{w}$ are the positive solutions of equations.

$$a_1 a_3 \hat{z}^{m+1} + a_1 a_3 \hat{z} - AK = 0 \quad and$$

$$a_2 a_4 \hat{w}^{m+1} - \hat{x}\hat{w}^m + a_2 a_4 \hat{w} - A - \hat{x} = 0 \qquad 6.6$$

Using Descartes rule of sign, (6.6) has only one positive root $\hat{z}$, can have either one or three positive roots depending on the parameter. Consider (6.6),

$$a_2 a_4 \hat{w} - \hat{x} = \frac{A}{1+\hat{w}^m} \tag{6.7}$$

$$f_1(\hat{w}) = a_2 a_4 \hat{w} - \hat{x}, \tag{6.8}$$

And R.H.S as

$$f_2(\hat{w}) = \frac{A}{1+\hat{w}^m} \tag{6.9}$$

As shown in Reboxetine or Placebo, $f_1(\hat{w})$ increase from –x to ∞ while $f_2(\hat{w})$ decreases, since both $f_1(\hat{w})$ and $f_2(\hat{w})$ are strictly monotone. Therefore, $\hat{E}(\hat{x},\hat{y},\hat{z},\hat{w})$ is unique for any m.

6.3.1 Stability

The investigated stability of the steady sate $\hat{E}(\hat{x},\hat{y},\hat{z},\hat{w})$ by linearization is $x = x' - \hat{x}, y = y' - \hat{y}, z = z' - \hat{z}, w = w' - \hat{w}$.

Then the linearized form of the system

$$\begin{aligned}
\dot{x}(t) &= -a_1 x(t) + \frac{Am\hat{z}^{m-1}}{(1+\hat{z}^m)^2} z(t-\Upsilon_1),\\
\dot{y}(t) &= -a_2 y(t) + x(t-\Upsilon_2) + \frac{Am\hat{w}^{m-1}}{(1+\hat{w}^m)^2} w(t-\Upsilon_2),\\
\dot{z}(t) &= -a_3 z(t) + Ky(t-\Upsilon_3),\\
\dot{w}(t) &= -a_4 w(t) + y(t-\Upsilon_4).
\end{aligned} \tag{6.10}$$

Let $\frac{Am\hat{z}^{m-1}}{(1+\hat{z}^m)^2}=\delta_1, \frac{Am\hat{w}^{m-1}}{(1+\hat{w}^m)^2}=\delta_2$

The characteristic equation is

$$D(\lambda,\Upsilon_i)=\lambda^4+A\lambda^3+\left(B+\delta_2 e^{-\lambda(r_3+r_5)}+\delta_1 K e^{-\lambda(r_1+r_4)}\right)\lambda^2+$$
$$\left(C+(a_1+a_3)\delta_2 e^{-\lambda(r_3+r_5)}+\delta_1 K(a_2+a_4)e^{-\lambda(r_1+r_4)}\right)\lambda+D+aa\delta_2 e^{-(r_3+r_5)}$$

$$+aa\delta_1 K e^{-(r_1+r_4)}+\delta_1 K\delta_2 e^{-(r_1+r_3+r_4+r_5)}=0 \qquad 6.11$$

When delays are absent in the system, the characteristic equation becomes,

$$D(\lambda,0)=\lambda^4+A\lambda^3+\left(B+\delta_2+\delta_1 K\right)\lambda^2+\left(C+(a_1+a_3)\delta_2+\delta_1 K(a_2+a_4)\right)\lambda$$

$$+D+aa\delta_2++aa\delta_1 K+\delta_1 K\delta_2=0, \qquad 6.12$$

Where

$$A=a_1+a_2+a_3+a_4,$$

$$B=a_1a_2+a_2a_3+a_3a_4+a_1a_4+a_1a_3+a_2a_4,$$

$$C=a_1a_2a_3+a_1a_2a_4+a_1a_3a_4+a_2a_3a_4, \qquad 6.13$$

$$D=a_1a_2a_3a_4.$$

The necessary and sufficient conditions for stability are given by Routh-Hurwitz condition, which are

$$D_1 = a_1 + a_2 + a_3 + a_4 > 0$$

$$D_2 = A\left(B + \delta_2 + \delta_1 K\right) - \left(C + (a_1 + a_3)\delta_2 + \delta_1 K(a_2 + a_4)\right) > 0$$

$$D_3 = \left(C + (a_1 + a_3)\delta_2 + \delta_1 K(a_2 + a_4)\right) D_2 > 0 \qquad 6.14$$

$$D_4 = DD_3 > 0$$

Since all the D_1, D_2, D_3, D_4 are positive, $D(\lambda, 0)$ has roots whose real parts are all negative. Hence the steady state is always stable.

To get the estimate on the lengh of delay (nearly 30min) is Reboxetine or Placebo. (a) reboxetine: COR 127893.20 ± 8125.75 nmol/l × min; ACTH2385.68 ± 387.19 pmol/l × min; GnRH 63026.59 ± 17594.68 pmol/l × min; PRL 113961.60 ± 10280.44pmol/l × min; (b) placebo: COR 83672.19 ± 5225.20 nmol/l × min; ACTH 1449.83 ± 190.67pmol/l × min; GnRH 11008.34± 5402.34 pmol/l × min; PRL 64663.28 ± 7283.62 pmol/l × min). the Nyquist criterion is used for solutions**[141].**

6.3.2 Estimation for the length of delay using Laplace transform

$$\dot{x}(t) = -a_1' x(t) + \delta_1' z^m(t - \Upsilon_1),$$

$$\dot{y}(t) = -a_2' y(t) + x(t - \Upsilon_2) + \delta_2' w^m(t - \Upsilon_3),$$

$$\dot{z}(t) = -a_3' z(t) + K y(t - \Upsilon_3), \qquad 6.15$$

$$\dot{w}(t) = -a_4' w(t) + y(t - \Upsilon_4),$$

Let $\bar{x}(t), \bar{y}(t), \bar{z}(t), and\ \bar{w}(t)$ denote the Laplace transform of $x(t), y(t), z(t), and\ w(t)$ respectively. The Laplace transform of the system (6.14) yields.

$$(Q - a_1')\bar{x}(Q) = \delta_1' e^{-Qr_1}\bar{z}(Q) + \delta_1' e^{-Qr_1} K_1(Q) + x(0)$$

$$(Q - a_2')\bar{y}(Q) = e^{-Qr_2}\bar{z}(Q) + e^{-Qr_3} K_1(Q) + \delta_1' e^{-Qr_2}\bar{w}(Q) + \delta_1' e^{-Qr_3} K_3(Q) + y(0)$$

$$(Q - a_3')\bar{z}(Q) = K e^{-Qr_4}\bar{z}(Q) + K e^{-Qr_4} K_1(Q) + z(0)$$

$$(Q - a_4')\bar{w}(Q) = e^{-Qr_5}\bar{y}(Q) + e^{-Qr_5} K_5(Q) + w(0) \qquad 6.16$$

$$where\quad K_1(Q) = \int_{-r_1}^{\infty} e^{-Qt} z(t)dt,\ K_2(Q) = \int_{-r_2}^{\infty} e^{-Qt} x(t)dt,\quad K_3(Q) = \int_{-r_3}^{\infty} e^{-Qt} w(t)dt,$$

$$K_4(Q) = \int_{-r_4}^{\infty} e^{-Qt} z(t)dt,\ K_5(Q) = \int_{-r_5}^{\infty} e^{-Qt} z(t)dt,$$

Rearranging (6.10),

$$(Q^4 + AQ^3 + \left(B + \delta_2 e^{-Q(r_3+r_5)} + \delta_1 K e^{-Q(r_1+r_4)}\right)Q^2 + \left(C + (a_1+a_3)\delta_2 e^{-Q(r_3+r_5)} + \delta_1 K(a_2+a_4)e^{-Q(r_1+r_4)}\right)Q$$
$$+(D + a_2 a_3 \delta_1 e^{-(r_3+r_5)} + a_1 a_3 \delta_2 K e^{-(r_3+r_5)} + \delta_1 K \delta_2 e^{-(r_1+r_3+r_4+r_5)})\bar{y}(Q)$$

$$= x_0(Q+a_3)(Q+a_4)e^{-Qr_2} + y_0\left\{(Q+a_1)(Q+a_3)(Q+a_4) + (Q+a_4)\delta_1 K e^{-Q(r_1+r_4)}\right\} - \qquad 6.17$$

$$z_0(Q+a_4)\delta_1 e^{-Q(r_1+r_2)} - w_0\left\{(Q+a_1)(Q+a_3)\delta_2 e^{-Qr_3} + K\delta_1\delta_2 e^{-Q(r_1+r_2+r_3+r_4)}\right\}$$

$$-(Q+a_3)(Q+a_4)\delta_1 e^{-Q(r_1+r_4)}K_1(Q) + \left\{(Q+a_1)(Q+a_3)(Q+a_4)e^{-Qr_2} + (Q+a_4)\delta_1 K e^{-Q(r_1+r_2+r_4)}\right\}K_2(Q)$$
$$-\left\{(Q+a_1)(Q+a_3)(Q+a_4)e^{-Qr_3} + (Q+a_4)\delta_1 K e^{-Q(r_1+r_3+r_4)}\right\}K_3(Q)$$
$$-(Q+a_4)K\delta_1 e^{-Q(r_1+r_3+r_4)}K_4(Q) - \left\{(Q+a_1)(Q+a_3)\delta_2 e^{-Q(r_3+r_5)} + K\delta_1\delta_2 e^{-Q(r_1+r_2+r_3+r_5)}\right\}K_5(Q)$$

$$= x_0\left\{Q^2 + (a_2+a_4)Q + a_3a_4\right\}e^{-Qr_2}$$
$$+y_0\left\{Q^3 + (a_1+a_3+a_4)Q^2 + (a_1a_4 + a_1a_3 + a_3a_4)Q + (Q+a_4)K\delta_1 e^{-Q(r_1+r_4)} + a_1a_3a_4\right\}$$
$$-z_0(Q+a_4)\delta_2 e^{-Q(r_1+r_2)} - w_0\left\{(Q^2 + (a_1+a_3)Q + a_1a_3)\delta_2 e^{-Q(r_3)} + K\delta_1\delta_2 e^{-Q(r_1+r_3+r_4)}\right\}$$

$$-\left\{Q^2+(a_1+a_4)Q+a_3a_4\right\}\left\{Q^3+(a_1+a_3+a_4)Q^2+(a_1a_4+a_1a_3+a_3a_4)Q+(Q+a_4)K\delta_1 e^{-Q(r_1+r_4)}+a_1a_3a_4\right\}e^{-Q(r_1+r_2)}K_1(Q)$$

$$+z\left[\left\{Q^3+(a_1+a_3+a_4)Q^2+(a_1a_4+a_1a_3+a_3a_4)Q+a_1a_3a_4\right\}e^{-Qr_3}+(Q+a_4)\delta_1 K e^{-Q(r_1+r_2+r_4)}\right]K_2(Q)$$

$$-\left[\left\{Q^3+(a_1+a_3+a_4)Q^2+(a_1a_4+a_1a_3+a_3a_4)Q+(Q+a_4)K\delta_1 e^{-Q(r_1+r_4)}+a_1a_3a_4\right\}\delta_2 e^{-Q(r_3)}+(Q+a_4)K\delta_1\delta_2 e^{-Q(r_1+r_2+r_4)}\right]K_3(Q)$$

$$-(Q+a_4)K\delta_1 e^{-Q(r_1+r_3+r_4)}K_4(Q)$$
$$-\left\{Q^2+(a_2+a_4)Q+a_1a_3\delta_2 e^{-Q(r_1+r_4)}+K\delta_1\delta_2 e^{-Q(r_1+r_3+r_4+r_5)}\right\}K_5(Q).$$

The inverse Laplace transform of $\bar{y}(Q)$ will have which exponentially increase with time, it $\bar{y}(Q)$ has poles with positive real parts. The conditions for

stability of $\hat{E}(\hat{x},\hat{y},\hat{z},\hat{w})$ are given by

$$\mathrm{Im}\Delta(iv_0) > 0, \tag{6.18}$$

$$\mathrm{Re}\,\Delta(iv_0) = 0, \tag{6.19}$$

Where

$$\Delta(Q) = \begin{bmatrix} Q^4 + AQ^3 + \left(B + \delta_2 e^{-Q(r_3+r_5)} + \delta_1 K e^{-Q(r_1+r_4)}\right)Q^2 + \\ \left(C + (a_1+a_3)\delta_2 e^{-Q(r_3+r_5)} + \delta_1 K(a_2+a_4)e^{-Q(r_1+r_4)}\right)Q \\ +(D + a_2a_3\delta_1 e^{-(r_3+r_5)} + a_1a_3\delta_2 K e^{-Q(r_3+r_5)} + \delta_1 K\delta_2 e^{-Q(r_1+r_3+r_4+r_5)}) \end{bmatrix} \tag{6.20}$$

and v_0 is the smallest positive root of the equation (6.18), conditions(6.19) and(6.18) in this case become

$$\begin{aligned} &v_0^4 - Bv_0^2 + D = K\delta_1 v_0^2\cos(T_1v_0) + \delta_2 v_0^2\cos(T_2v_0) - K\delta_1 v_0(a_2+a_4)\sin(T_1v_0) \\ &-\delta_2 v_0(a_1+a_3)\sin(T_2v_0) - K\delta_1 a_2a_4\cos(T_1v_0) - \delta_2 a_1a_3\cos(T_1v_0) - K\delta_1\delta_2\cos(T_1+T_2)v_0 \end{aligned} \tag{6.21}$$

$$\begin{aligned} &-Av_0^3 + Cv_0 > -K\delta_1 v_0^2 sin(T_1v_0) - \delta_2 v_0^2\sin(T_2v_0) - K\delta_1 v_0(a_2+a_4)\cos(T_1v_0) \\ &\delta_2 v_0(a_1+a_3)\sin(T_2v_0) - K\delta_1 a_2a_4\cos(T_1v_0) - \delta_2 a_1a_3\sin(T_1v_0) - K\delta_1\delta_2\sin(T_1+T_2)v_0. \end{aligned} \tag{6.22}$$

To estimate the length of delay the following conditions are utilized

$$v^4 - Bv^2 + D = K\delta_1 v^2\cos(T_1v) + \delta_2 v^2\cos(T_2v) - K\delta_1 v(a_2+a_4)\sin(T_1v)$$

$$-\delta_2 v(a_1+a_3)\sin(T_2 v)-K\delta_1 a_2 a_4\cos(T_1 v)-\delta_2 a_1 a_3\cos(T_1 v)-K\delta_1\delta_2\cos(T_1+T_2)v, \qquad 6.23$$

$$-Av^3+Cv>-K\delta_1 v^2 sin(T_1 v)-\delta_2 v^2\sin(T_2 v)-K\delta_1 v(a_2+a_4)\cos(T_1 v)$$

$$\delta_2 v(a_1+a_3)\sin(T_2 v)-K\delta_1 a_2 a_4\cos(T_1 v)-\delta_2 a_1 a_3\sin(T_1 v)-K\delta_1\delta_2\sin(T_1+T_2)v. \qquad 6.24$$

Recall that $\hat{E}$ will be stable if the inequality (6.24) holds at $v=v_0$, when v_0 is the first positive root of equation (6.21)

Here v_+ is always greater than or equal to v_0.

$$v^4-v^2(B+K\delta_1+\delta_2)-v\left[K\delta_1(a_2+a_4)+\delta_2(a_2+a_4)\right]+ \left[D-K\delta_1 a_2 a_4-\delta_2 a_1 a_3+K\delta_1\delta_2\right]=0, \qquad 6.25$$

$$v_+=\sqrt{p}+\sqrt{q}-\frac{G}{\sqrt{p}\sqrt{q}} \qquad 6.26$$

where

$$p+q+r=\frac{B+K\delta_1+\delta_2}{2},$$

$$pq+qr+rp=\left(\frac{B+K\delta_1+\delta_2}{4}\right)^2-\left[D-K\delta_1 a_2 a_4-\delta_2 a_1 a_3+K\delta_1\delta_2\right],$$

$$\sqrt{p}\sqrt{q}\sqrt{r}=\frac{K\delta_1(a_2+a_4)+\delta_2(a_2+a_4)}{8}=-\frac{G}{2}.$$

Here v_+ is always independent of T_1 and T_2. The estimate on T_1 and T_2 is

$$Av^2 < C + \left(K\delta_1 v - \frac{K\delta_1 a_2 a_4}{v} \right) \sin(T_1 v) + \left(\delta_2 v - \frac{\delta_2 a_1 a_3}{v} \right) \sin(T_2 v) \qquad 6.27$$

$$+ K\delta_1 (a_2 + a_4) \cos(T_1 v) + \delta_2 (a_2 + a_4) \cos(T_2 v) - \frac{K\delta_1 \delta_2}{v} \sin(T_1 + T_2) v. \qquad 6.28$$

Note that when Υ_i=0, the solution

$$v^2 = \frac{B + K\delta_1 + \delta_2 \pm \left(\left[B + K\delta_1 + \delta_2 \right]^2 - 4\left[D - K\delta_1 a_2 a_4 - \delta_2 a_1 a_3 + K\delta_1 \delta_2 \right] \right)^{\frac{1}{2}}}{2} \qquad 6.29$$

$$\Rightarrow Av^2 = \frac{L}{2} \pm \sqrt{\left(\frac{L}{2} \right)^2 - A^2 M},$$

Where,

$$(i)\ L = A(B + K\delta_1 + \delta_2),$$

$$(ii)\ M = (D + K\delta_1 a_2 a_4 + \delta_2 a_1 a_3 + K\delta_1 \delta_2), \qquad 6.30$$

$$(iii)\ N = K\delta_1 (a_2 + a_4) + \delta_2 (a_2 + a_3) + C,$$

$$(iv)\ S = AB - C + K\delta_1 (a_2 + a_3) + \delta_2 (a_2 + a_4).$$

Now, since from (6.28)

$$D_3 = \left\{ K\delta_1 (a_2 + a_4) + \delta_2 (a_1 + a_3) + C \right\} \left[AB - C + K\delta_1 (a_1 + a_3) + \delta_2 (a_2 + a_4) \right]$$

$$-A^2 (D - K\delta_1 a_2 a_4 + \delta_2 a_1 a_3 + K\delta_1 \delta_2) > 0, \ \ i.e., SN > A^2 M.$$

So,

$$Av^2 < C + K\delta_1(a_2 + a_4) + \delta_2(a_1 + a_3), \qquad 6.31$$

(6.28) is valid for T_i=0 at $v = v_0$, Now by continuity it will hold for small T_i>0 at $v = v_0$

Substituting v^2 from (6.23) into (6.24),

$$\left[A(K\delta_1 v^2 - K\delta_1 a_2 a_4) - K\delta_1(a_2 + a_4)v^2\right]\cos T_1 v + \left[K\delta_1 a_2 a_4 v - K\delta_1 v^3 - AK\delta_1 v(a_2 + a_4)\right]\sin T_1 v$$

$$+\left[A(\delta_2 v^2 - \delta_2 a_1 a_3) - \delta_2(a_1 + a_3)v^2\right]\cos T_2 v + \left[\delta_2 a_1 a_3 v - \delta_2 v^3 - A\delta_2 v(a_1 + a_3)\right]\sin T_2 v$$

$$+K\delta_1\delta_2 v\sin(T_1 + T_2)v - AK\delta_1\delta_2\cos(T_1 + T_2)v < (C - AB)v^2 + AD,$$

$$\left[A(K\delta_1 v^2 - K\delta_1 a_2 a_4) - K\delta_1(a_2 + a_4)v^2\right](\cos T_1 v - 1) + \left[K\delta_1 a_2 a_4 v - K\delta_1 v^3 - AK\delta_1 v(a_2 + a_4)\right]\sin T_1 v$$

$$+\left[A(\delta_2 v^2 - \delta_2 a_1 a_3) - \delta_2(a_1 + a_3)v^2\right](\cos T_2 v - 1) + \left[\delta_2 a_1 a_3 v - \delta_2 v^3 - A\delta_2 v(a_1 + a_3)\right]\sin T_2 v$$

$$+K\delta_1\delta_2 v\sin(T_1 + T_2)v - AK\delta_1\delta_2\cos((T_1 + T_2)v - 1) < \begin{bmatrix} A(D + K\delta_1 a_2 a_4 + \delta_2 a_1 a_3 + K\delta_1\delta_2) - \\ v^2\{A(BK\delta_1 + \delta_2) + (C + K\delta_1(a_2 + a_3) + \delta_2(a_2 + a_4))\} \end{bmatrix}$$

6.32

$$= AM - v^2 D_2 < AM \equiv \zeta.$$

Denote the L.H.S of (6.32) as $\Omega_1(T_1, v) + \Omega_2(T_2, v)$, using the estimates $\sin T_i v \le T_i v$

$$\sin(T_1+T_2)v \le (T_1+T_2)v,$$

$$(1-\cos T_i v) \le \frac{1}{2}T_i^2 v^2$$

$$1-\cos(T_1+T_2)v \le (T_1^2+T_2^2)v^2,$$

$$\Omega_1(T_1,v)+\Omega_2(T_2,v) \le \varphi_1(T_1,v)+\varphi_2(T_2,v)$$

$$\equiv\left[K\delta_1 v^2(\frac{1}{2}\left|v^2(a_1+a_2)-Aa_2a_4\right|+AK\delta_1)T_1^2+K\delta_1 v^2\left(v^2+A(a_2+a_4\right)-a_2a_4+\delta_2)T_1\right]+$$
$$\left[\delta_1 v^2(\frac{1}{2}\left|v^2(a_2+a_4)-Aa_1a_3\right|+AK\delta_1)T_2^2+\delta_2 v^2\left(v^2+A(a_1+a_3\right)-a_1a_3+K\delta_1)T_2\right]<\zeta \qquad 6.33$$

Note that for $0\le v\le v_+$,

$$\Omega_1(T_1,v)+\Omega_2(T_2,v) \le \varphi_1(T_1,v)+\varphi_2(T_2,v) \le \varphi_1(T_1,v_+)+\varphi_2(T_2,v_+),$$
$$\varphi_1(T_1,v_+)+\varphi_2(T_2,v_+)<\zeta,\ \Omega_1(T_1,v_0)<\zeta,\quad \Omega_2(T_2,v_0)<\zeta \qquad 6.34$$

Let $T_1^+ and T_2^+$ denote the unique root of

$$\varphi_1(T_1,v_+)=\alpha\zeta,\ \varphi_2(T_2,v_+)=\beta\zeta,\ \alpha+\beta=1$$

$$T_1^+=\frac{1}{2M_1}\left[-N_1+\sqrt{N_1^2+4\alpha\zeta M_1}\right],$$

$$T_2^+=\frac{1}{2M_2}\left[-N_2+\sqrt{N_2^2+4\alpha\zeta M_2}\right],$$

$$M_1=K\delta_1 v_+^{\ 2}(\frac{1}{2}\left|v_+^{\ 2}(a_1+a_3)-Aa_2a_4\right|+AK\delta_1)$$

$$N_1=K\delta_1 v_+^{\ 2}\left(v_+^{\ 2}+A(a_2+a_4\right)-a_2a_4+\delta_2)$$

$$M_2 = \delta_1 v_+^{\,2}\left(\frac{1}{2}\left|v_+^{\,2}(a_2 + a_4) - Aa_1a_3\right| + AK\delta_1\right)$$

$$N_2 = \delta_2 v_+^{\,2}\left(v_+^{\,2} + A(a_1 + a_3\right) - a_1a_3 + K\delta_1)$$

6.35

Then for $T_1 < T_1^+$ and $T_2 < T_2^+$. The Nyquist criterion holds give the estimates for the length of delay for which stability of Reboxetine or Placebo. a) reboxetine: COR 127893.20 ± 8125.75 nmol/l × min; ACTH2385.68 ± 387.19 pmol/l × min; GnRH 63026.59 ± 17594.68 pmol/l × min; PRL 113961.60 ± 10280.44pmol/l × min; (b) placebo: COR 83672.19 ± 5225.20 nmol/l × min; ACTH 1449.83 ± 190.67pmol/l × min; GnRH 11008.39 ± 5402.34 pmol/l × min; PRL 64663.28 ± 7283.62 pmol/l × min) **[97,127].**

6.4 Data analysis

The descriptive and graphical evaluation of the mean curves (hormonal concentrations, MAP, heart rate), the areas under the curve,(0 to 420 minutes) were calculated according to the fourth order Runge Kutta Feldberg method, for obtaining accurate numerical results, representing the total COR, ACTH, GnRH, and PRL secretion and the following oral administration of placebo or reboxetine (4 mg).

6.5 Results

The program was developed by using the fourth order Runge Kutta Fehlberg method **[141]** for obtaining accurate numerical results and constructed following algorithm to incorporate delay terms. The numerical results from computer simulation have been given as Figures 6.5.1-6.5.4, which are been discussed.

So, the theoretical value is in good agreement with the experimental results. Physiological observation regarding the correlation of COR, ACTH, GnRH and PRL is clearly depicted in Figure 6.5.1-6.5.4. The values of parameters were chosen partly to be physiologically reasonable, partly to fit existing human data. The figures for the delays are based on the data, the values of $\alpha, \beta, \delta, \gamma$ were taken from **[141]**.

6.5.1 COR and ACTH secretion

Initially (t = 0 min up to t = 420min) a considerable decrease of serum COR levels and plasma ACTH concentrations occurred which was comparable between the reboxetine condition and the placebo condition (Fig. 6.5.1) remarkable increase of serum COR and plasma ACTH concentrations happened.

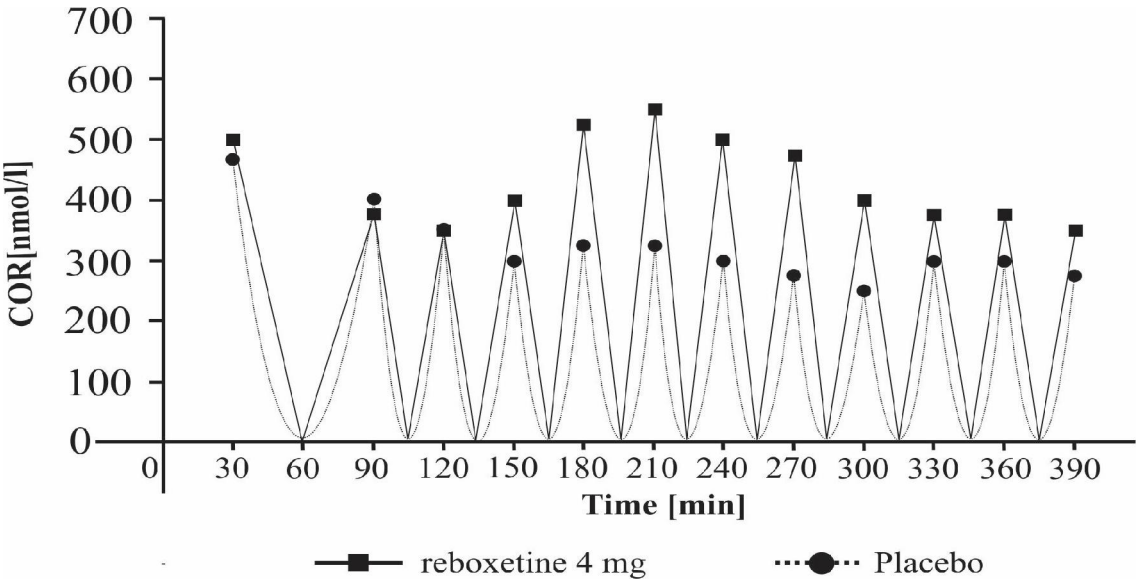

Figure. 6.5.1 Cortisol concentrations after 4 mg reboxetine and placebo in 12 healthy male subjects

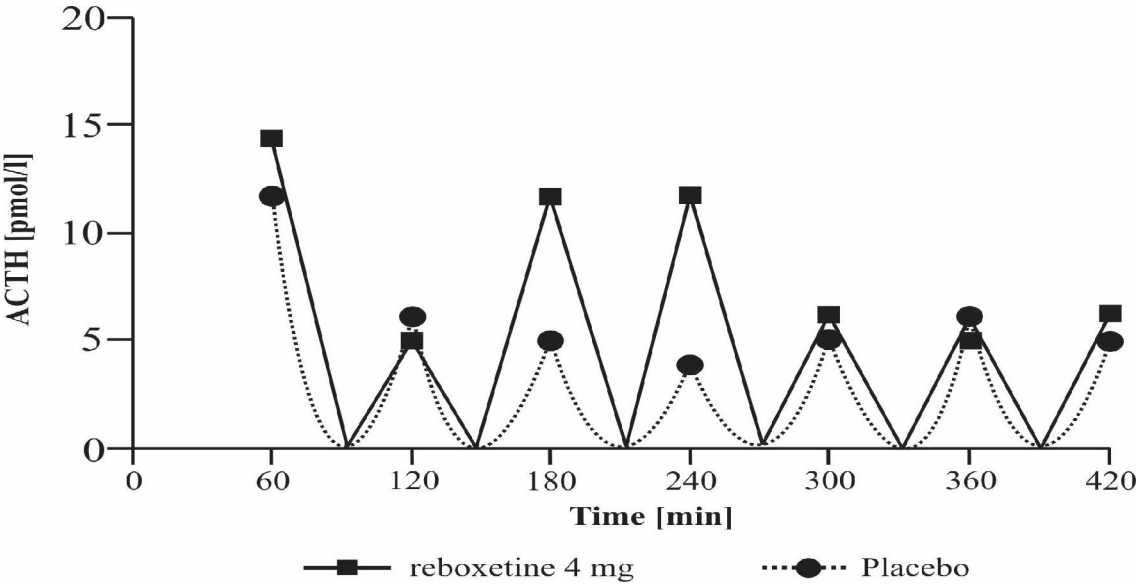

Figure. 6.5.2 ACTH concentrations after 4 mg reboxetine and placebo in 12 healthy male subjects.

During the following time of the measurement period (t = 0 min up to t = 300 min), in the placebo condition a further slight reduction of serum COR and plasma ACTH concentrations could be seen reaching values below 250 nmol/l (COR) and below 5 pmol/l (ACTH). However, after reboxetine (4 mg PO), a The mean peak concentrations for COR and ACTH were found at t = 120 min and were 561.20 nmol/l and 12.20 pmol/l respectively.

Thereafter, the mean COR and ACTH levels dropped again and were comparable to those observed after placebo at the end of the measurement period. Both the mean serum COR area under curve (AUC) value and the mean plasma ACTH area under curve value were considerably enhanced in the reboxetine group (mean COR area under curve : 127893.20 ± 8125.75 0- 300 nmol/l × min; mean ACTH area under curve : 2385.68 ± 387.19 pmol/l × min) compared to the 0- 300 placebo group (mean COR area under curve : 83672.19 ± 5225.20 nmol/l × min; mean ACTH 0- 300 AUC : 1449.83 ± 190.67 pmol/l × min). The mathematical model indicated significantly higher COR area under curve and ACTH area under curve(AUC) values after administration of reboxetine compared to placebo (COR AUC : ACTH AUC :).

6.5.2 GnRH Secretion

Before administration of placebo or reboxetine, most GnRH concentrations measured were clearly below 200 pmol/l in both treatment groups (reboxetine, placebo). However, one healthy subject displayed a GnRH peak up to 521.31 pmol/l at $t = 0$ min during the placebo condition. The same volunteer also showed a considerable GnRH peak 30 minutes after administration of reboxetine (2007.53 pmol/l) thereby causing a moderate elevation of the mean value graph concerning the reboxetine condition at $t = 30$ min (Fig. 6.5.3).

Thereafter, this GnRH levels during reboxetine condition dropped to 264.06 pmol/l ($t = 360$ min) and increased again up to 1066.26 pmol/l ($t = 210$ min). In two further volunteers, slight and short-lasting GnRH elevations were found during placebo condition at $t = 240$ min (531.77 pmol/l) and $t = 360$ min (282.24 pmol/l). In all other subjects, GnRH levels remained lower than 200 pmol/l after placebo and showed a pronounced peak 120 minutes after administration of 4 mg reboxetine (Figure.6.5.3).

GnRH AUC values were 56026.59 ± 15594.87 pmol/l × min after reboxetine and11008.34± 5402.34 pmol/l × min after placebo. Using mathematical model, the difference reached statistical significance **[66, 67,136].**

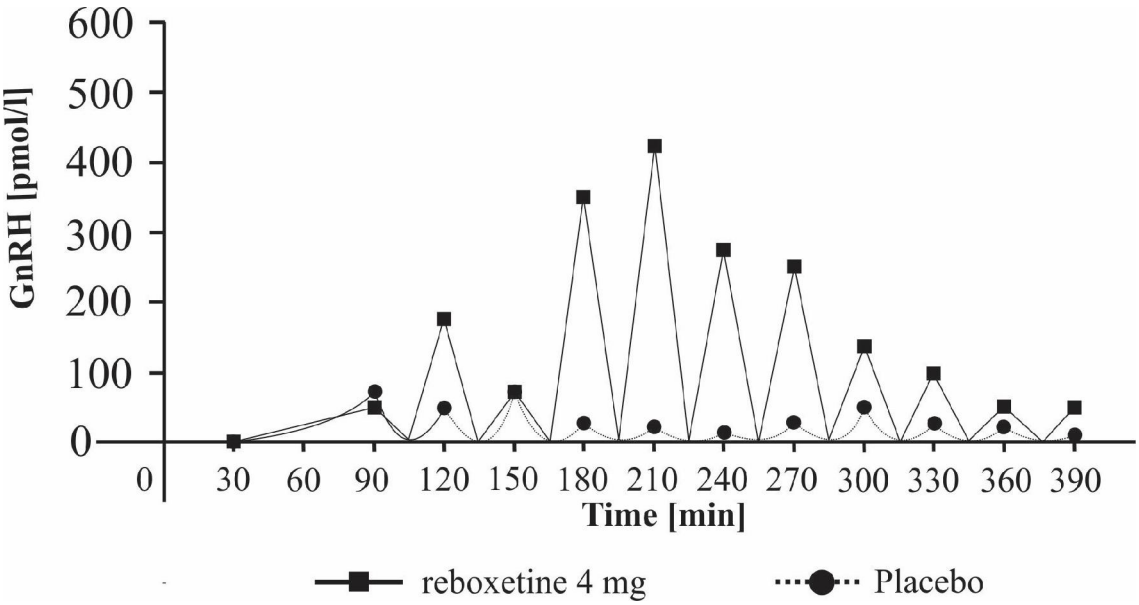

Figure.6.5. 3 GnRH concentrations after 4 mg reboxetine and placebo in 12 healthy male subjects.

6.5.3 PRL secretion

Between t = 60 min and t = 150 min, a moderate decrease was observed during both treatment conditions (placebo, reboxetine). When placebo was given, mean PRL levels further decreased up to the end of the measurement period. However, after administration Figure.6.5.4. prolactin concentrations after 4 mg reboxetine and placebo in 12 healthy male subjects of reboxetine (4 mg), a marked increase of mean PRL concentrations occurred reaching maximum levels between t = 150 min and t = 240 min (Figure 6.5.4) PRL.

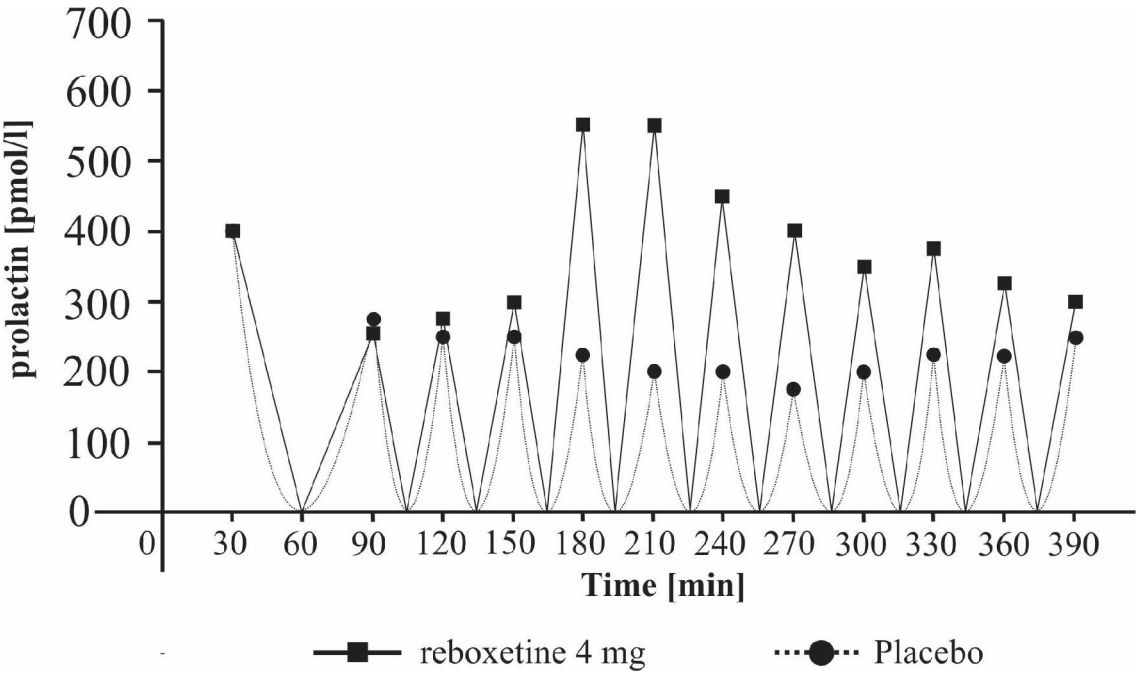

Figure 6.5.4

Using Mathematical Model area under curve values were significantly higher after reboxetine (113961.6 ± 10280.44 0 -3 00 pmol/l × min) than after placebo (64663.28 ± 7283.62 pmol/l × min).

6.6 Discussion

In the present study in healthy male subjects the selective NA reuptake inhibitor reboxetine exerted pronounced stimulatory effects on (i) COR, ACTH,(ii) GH and(iii) PRL secretion patterns. (i) COR and ACTH secretion regarding these data, one can assume that the increased ACTH and COR release after administration of reboxetine observed in study is caused by selective NA reuptake inhibition thereby enhancing NA concentrations in the synaptic cleft and stimulating CRH and/or vasopressin output via hypothalamic α_1 - adrenoceptors. The mathematical model indicated significantly higher COR AUC and ACTH AUC values after administration of reboxetine compared to placebo(ii) GnRH investigation can probably be put down to the fact that GnRH shows pulsatile secretion patterns with spontaneous elevation of GnRH concentrations up to 900pmol/l. This Mathematical model shows the difference reached statistical significance (iii) there are some convincing data suggesting a stimulatory role of hypothalamic adrenoceptors in PRL release in man remain unclear. This Mathematical Model area under curve values were significantly higher after reboxetine than after placebo.