COMPUTER-BASED STUDIES ON BIOPROCESS ENGINEERING

II - TOOLS FOR PROCESS OPERATION

S. FEYO DE AZEVEDO, P. PIMENTA, F. OLIVEIRA
Departamento de Engenharia Química
Faculdade de Engenharia da Universidade do Porto
Rua dos Bragas 4099 Porto Codex Portugal

E. C. FERREIRA
Departamento de Engenharia Biológica
Universidade do Minho
Largo do Paço 4719 Braga Codex Portugal

ABSTRACT

In this paper we review recent advances on the practice and theory of process control with particular emphasis to the operation of bioreactors. We present in detail a case-study on the modelling, model-based identification and adaptive control of fed-batch baker's yeast fermentation.

1. Practice and Theory of Process Control - An Overview

1.1. INTRODUCTION

After about thirty years of research and development digital systems and related methodologies have achieved the definite recognition of being vital tools for process operation, both in chemical and biochemical plants.

A kind of landmark in the history of process control can be recognised as having been reached at the close of the eighties. Nowadays, a large number of suppliers offer the capabilities of the digital technology, employing open architecture and standard operating systems and allowing external programming with high level languages.

For years researchers developed ideas and methods which as yet found little application in the industry. Much because these methods require computer and programming power which were not available in the industrial control equipment of the recent past.

A.R. Moreira and K.K. Wallace (eds.), Computer and Information Science Applications in Bioprocess Engineering, 27-49.

From the Smith predictor, through the state estimators to the more recent model based predictive control techniques, including also more and more improved PID controllers, methods can now be implemented at industrial scale.

The job for the future, and we shall assist to that, will be to incorporate new methodologies, some already existing and some still emerging, into those industrial equipment and thus bring such methodologies into operation.

The pace of progress depends on factors which go beyond research. It is worth to examine how the present stage has been reached in order to foresee the difficulties of the job ahead.

1.2. PROGRESS IN DIGITAL CONTROL TECHNOLOGY

The advancements in digital technology already had a most significant impact in the instrumentation and control industry.

Sensors and analytical instruments are the primary elements for process monitoring. Twork and Yacynych (1990) edited a review book on such equipment for bioprocess control. Sensors more and more employ digital technology, particularly for digital detection (e.g. - optical detection for refractometry-based sensors). Most analytical instruments have their performances improved by incorporating a microcomputer as a basic part of the equipment. Both sensors and instruments readily communicate with the control equipment via standard analogue signals or digital protocols. Yet, much research is necessary, particularly in the search for non-invasive sensors to operate on-line and in real-time. Just an example, reliable measurement of biomass has not yet been achieved.

In what concerns control systems, we have now reached the point where commercial, robust, reasonably simple to operate, computer-based control systems are available with the necessary characteristics to implement applications, on-line and in real-time. Most systems run a distributed philosophy with a central supervisory control computer linked via a digital communications data network to local modules. These can be analogue and digital input/output cards or local controllers (including programmable logic controllers) with their own I/O interfaces. A major improvement over older systems is on the large capacity to perform process monitoring (with reporting and alarm action) and to perform complex sequential control. Regulatory control is in most cases still limited to single input, single output (SISO) discrete PID, to discrete cascade control, to ratio control and to some limited feedforward control with lead-lag actions. This is a clear 'under utilisation' of the technology.

The 'step forward' which we observe in the existing commercial control systems is mainly related to the 'process control computer'. For some years the systems were practically closed to external programming in high level languages. Now, with the advances in hardware and operating systems, manufacturers are turning to open systems, employing standard hardware and software.

We find large scale applications of data acquisition and control typically based in VAX, SUN, HP or IBM stations (e. g., in oil refining, Hydrocarbon Processing, 1991). Many industrial systems are already based in microcomputers. This includes not only industrial data acquisition and monitoring systems, (e. g., IMPs from Schlumberger) but also process control systems (e.g. SCAP from SCAPE EUROPE, IA from Foxboro). SCAP runs under QNX and offers adaptive

predictive control (Martin Sanchez, 1976; Martin-Sanchez *et al.*, 1984). The IA system allows programming in the C language at the 'supervisory' level.

The trend observed above applies as well to expert systems. Computer Scientists developed artificial intelligence in large part around the computer language LISP (List Processing) and more recently around PROLOG. Both process symbols and list symbols. Special purpose computer platforms ('symbolic' machines) dedicated to symbolic processing were developed assuming that this would bring superior performances. But reviewing the availability of expert systems shells (Stock, 1989; Harmon, 1990) one concludes that traditional computers and more common languages are being employed. Recent work on 'continuous process improvement through induction and analogical learning' (Saraiva and Stephanopoulos, 1992) is being developed in a traditional mainframe with FORTRAN.

1.3. PROGRESS IN PROCESS CONTROL THEORY

Years ago Foss (1973), Lee and Weekman (1976) and Kestenbaum *et al.* (1976) analysed critically the theoretical developments and trends in process control. There was at the time a recognised gap between theory and practice. Rightly, the authors generally pointed that it was up to the theory to jump and cover the gap.

The algorithms employed were based on models which hardly represented real chemical and biochemical processes. They lacked the required robustness for industrial application. And they required a computer power which was not available in control systems. Very important and pragmatic developments both in computer systems (as already seen) and in process control theory occurred since those days. The criticism of the past was largely taken into account. Methods and algorithms were developed which specifically had in mind the characteristics of real processes, viz - non-linear, often multivariable without the possibility of decoupling, ...

Bequette (1991) extensively reviews contributions for the non-linear control of chemical processes. Cadman (1991) similarly addresses the problem of bioreactor control and Lübbert (1991) produces a comprehensive description of automation in biotechnology.

A number of topics emerge on the front-line of concern -

(i) the requisites for the use of on-line techniques and for the use of packages for process monitoring
(ii) the role of mathematical models in bioengineering, for -
- process analysis (discussed in part I)
- monitoring the behaviour of internal variables that are impossible to measure on-line
model based control
(iii) the problem of system identification both for stochastic and deterministic models
(iv) adaptive control, once again based on stochastic or deterministic modelling.

The problems concerning sensors and instrumentation in biotechnology are dealt with by Schügerl in his many papers (1988), including his lessons in this book. The required characteristics of packages for on-line monitoring will be discussed later in section 2.

Self tuning and adaptive control have been now for several years a reference for research and development. The current trend, which very much seems successful, is to couple a robust parameter and state estimation to a robust control algorithm.

Bastin, Dochain, Pomerleau and Perrier, published a string of papers describing model based software sensors, identification and adaptive control strategies for bioreactors (Dochain and Bastin, 1985; Bastin and Dochain, 1990; Dochain, 1991; Dochain *et al.* 1991; Dochain *et al.*, 1992; Pomerleau *et al.* , 1989; Pomerleau and Perrier, 1990; Pomerleau and Viel, 1992).

The general methodology on state and parameter estimation makes full use of the non-linear structures accepted for fermentation processes, with the particular feature that it avoids the need to know, a priori, kinetic parameters.

The concept of exact linearizing control is extensively detailed by Bastin and Dochain (1990) and will be presented in section 3 with reference to a case study with baker's yeast fermentation.

Model identification and adaptive control based on stochastic modelling of dynamical systems have as well received much attention of researchers. Beck and Young (1989) provide a very concise review on system identification and state and parameter estimation. The textbooks of Åström and Wittenmark (1984, 1989) and of Goodwin and Sin (1984) and the texts of Graupe (1987) and of Landau (1987) provide a background on concepts and theory.

In the last few years important progress has been achieved when, in independent developments, control strategies were proposed which couple methods of model based identification with the concept of controlling an a priori known process by making use of predictions of its future behaviour. Such methodology is known as Model Based Predictive Control (MBPC).

The first known version of MBPC is the adaptive predictive method (Martin Sanchez, 1976, 1986; Martin Sanchez *et al.*, 1984). Other versions are the extended-prediction self adaptive control (de Keyser and Van Canwenbergh, 1979), the generalised predictive control (Clarke *et al.*, 1987) and the MUSMAR (Mosca *et al.*, 1984). The theoretical basis, the practical advantages and the basic tools for building a MBPC strategy are clearly described by Richalet (1990), de Keyser (1990) and Clarke (1990, 1991).

Many successful industrial applications of MBPC are reported (de Keyser, 1990) and industrial control systems are now available which implement the methodology as a standard.

There are known draw-backs in the available theories and as such research goes on. An whole issue of Chemical Engineering Science (1992, Vol. 47, n. 4, 705-958) is dedicated to applications of model based control to chemical and biochemical engineering problems. Possibly the more relevant question is that MBPC requires a suitable process model. Such model must be able to represent adequately the process dynamics behaviour and yet must be simple enough not to preclude on-line identification. It often so happens that this is a difficult compromise to achieve.

Artificial neural networks represent alternative forms of representation of system dynamics which have been receiving much attention for the modelling of bioprocesses. The subject is discussed by Bungay (1992) in this book and is out of the scope of this paper. Yet a couple of

comments are appropriate. Morris *et al.* (1991) apply neural nets to fed-batch penicillin fermentation and to continuous mycelial fermentation; Cooney *et al.* (1991) discuss the general application to bioprocess operation; Rivera and Karim (1992) tested neural network predictions in a bioreactor for the production of ethanol; Simutis *et al.* (1992) have recently studied the use of this method in predicting the substrate degradation during a production scale beer brewery fermentation. The conclusion of the latter application was that neural nets provide results which are similar to those obtained with the use of extended Kalman filters based on dynamic mathematical process models, but requiring significantly less development time.

Fuzzy reasoning linked to knowledge bases is another emerging methodology. Lübbert *et al.* (1992) have applied with excellent results a fuzzy supported extended Kalman filter in the description of the beer fermentation process. The textbook by Kosko (1992) presents in detailed neural networks and fuzzy theory from an unified engineering perspective.

There is today a strong interest on the application of real time knowledge based systems (RTKBS) to the process industries. The interest is as 'obvious' as that on 'adaptive' strategies. Essentially RTKBS are a form of memorising experience and using it in all possible ways - fault detection, emergency action, programmed operation, model switching, etc. In daily plant routine operators run their processes by experience. They adapt operation (reference points) manually when facing situations which are not programmed but which are not strange to their 'know-how'. Or they may well have written recipes of multiple set point profiles to implement in batch and fed-batch operation. Application of RTKBS at supervisory level in a computer based control system seems to be the future 'computerized' response for operation.

Stephanopoulos and Stephanopoulos (1986) discussed requirements of RTKBS for application to bioprocesses. Morris *et al.* (1991) and Aynsley *et al.* (1990) report results of a study on the production of penicillin. More recently, at the 5th International Conference on Computer Applications in Fermentation Technology a paper was given of the activity towards integrating the 'G2' (Gensym Corporation) real-time expert system into a process control strategy for the control of industrial fermentation (Fowler *et al.*, 1992). This paper is singled out for what it represents of a link between research and practice but it is accepted that artificial intelligence, expert systems and neural networks are somehow still futuristic methods for practical industrial control.

A closing reference on AI and neural networks. As for other subjects, there is today a news group dedicated to neural nets in the USENET worldwide communications network (comp.ai.neural-nets). It aims to be an open forum for all aspects of neural networks. Since it is user driven, both general and specific topics can be discussed.

1.4. WHAT ABOUT PID CONTROL ?

At an international process control conference in 1991 it was stated that 90% of industrial control requirements are still met by proportional-integral-derivative (PID) type controllers (Deshpande, 1992). Richalet (1990) wrote recently that 'an impressive majority of problems can be solved by simple and factual PI controllers'.

It is clear that the justification for the so-called modern algorithms has to be found on the relatively few but economically significant processes which are not so appropriately controlled

by PID algorithms. This includes inverse-response, non-linear, integrating and multivariable processes.

Model based predictive control methods seem to have all the conditions to become future standards because they combine superior performance with reasonably accessible concepts to non-experts. But the simple to understand, simple to operate and reasonably efficient PID will stay in operation for many years. Looking back to its birth, fifty years ago, PID is probably the most successful concept of this century as far as process operation is of concern.

One should note that PID can well be coupled to identification stages and to model based software sensors.

For a number of years attempts have been made to improve the basic PID algorithms. Åström and Hagglund (1988) dedicate a book to summarise the state of the art of PI control and methods for the automatic tuning of such controllers. Autotuning is a concept related to adaptive control which was developed and put into commercial equipment. Examples of these are the Exact^{TM} system of Foxboro, the Statt Control Instruments Automation^{TM}, the Electromax V^{TM} from Leeds of Northamp and Turnball Controls Systems 6355 Autotuning Controller^{TM}.

Recently Omron launched a digital PID controller with internal feedforward for set point changes. Also they launched the first fuzzy feedback controller. Both types come with fixed sampling periods of 0.5 seconds which is reasonable for the vast majority of chemical and biochemical processes.

The interest of the search for improved PID action is confirmed by the work of Deshpande (1992) who proposes the Q-PID controller. It includes a lead-lag network and dead-time compensation. Digital controllers open the way to major improvements due to the flexibility of building-in new algorithms. In general, feedforward action is not taken as a standard but it normally improves significantly the behaviour of the feedback action. We believe that digital feedback-feedforward controllers (with two inputs and one output) based on the feedback PID concept would be a simple and effective control equipment.

1.5. STEPS IN SYSTEM DEVELOPMENT.

It should have emerged from this analysis that there are clear steps on the study and implementation of a control system, viz. -

i. Process monitoring - which includes data acquisition, alarms, safety on-off action and some basic data analysis.

ii. Data interpretation - which may be off-line or on-line parameter estimation and system identification.

iii. Optimisation and process control - where optimisation is usually carried out off-line, based on a deterministic representation of the process or the plant.

In the reminder of this paper we shall discuss some of our experience in industrial process monitoring and we shall present an application on model-based identification and control of a fed batch baker's yeast fermentation.

2. Process Monitoring

Process monitoring is the single application where computers can have a fast and significant impact in process operation. Production managers are usually more keen in knowing instantaneously what is happening in the plant than in having a sophisticated control.

Any medium and large scale commercial control system includes the tools for process monitoring but usually these systems are expensive and the decision on acquiring them is difficult to take.

There are available nowadays low cost hardware-software packages which aim primarily at process monitoring but also incorporate low level control capabilities. Such systems are PC-based, run under MS-DOS and have a distributed network of interface modules, with a protocol assuring communication through a single cable over distances above one kilometre.

Basic process monitoring should include scanning input, alarms and alarm action, historian report, graphical and spreadsheet-format display and finally storing data on disk. In practical terms the following general characteristics are expected from a process monitoring system:

i. Analog inputs - capability for scanning of current, voltage, resistance, temperature (thermocouple), temperature (RTD) and strain.
ii. Digital inputs - frequency, period, events and counts.
iii. Digital outputs - voltage free for alarm action and manual on-off control of external processes.
iv. Analog outputs - for limited regulatory control and for external communication.
v. System configuration - should display automatically all choices for each channel; should allow the set-up and saving of several alternative configurations for quick set-up of system operation.
vi. Headers - should allow reasonable identification of each channel.
vii. Conversions - should include linear and non-linear conversion. The latter should include polynomial, exponential (several types), hyperbolic and sinusoidal functions.
viii. Interchannel and individual calculations - for functions between input channels or for internal individual channel calculations such as averages, totalizers, maximums and minimums.
ix. Programmed timer - there would be the possibility of assigning different sampling and recording rates to individual or groups of channels.
x. Alarms - different types of alarm decisions based on level limits, rate of change limits or deviation limits. Alarms must be linked to digital output channels for on-off action.
xi. Display - a wide variety should be possible from spreadsheet-format to graphical display. The latter should include bargraph, trend plots and localized display in synoptic. Displays should be available in ´pages´.
xii. Synoptic - process drawing capabilities are required with definition of localized variable outputs.
xiii. Data recording and data printing - should be available by modules - process data, alarm and control action reports. It is particularly relevant that such data is recorded in a format directly compatible with spreadsheet packages.
xiv. Data communications - with other computers, using serial communication.

xv. Link with external subprograms - it is most convenient that software written by the user can be added to the package. With microcomputers this currently means C language and MS-Quick Basic. In the near future alternatives may be available.

Together with looking into the characteristics mentioned the potential buyer would do well in inquiring about a few more details such as - Is the package standard ? Is it available off-shelf ? For how many years is it in the market ? What are the limitations of hardware ? Is it fully PC-compatible ? Which requirements for graphical display ? What is the need and/or the flexibility of programming ? What is the available applications consultancy provided by the vendor and which technical support can we expect ? These two final questions are particularly relevant where the equipment comes from outside the buyer's home country.

3. A Case Study - Modelling, Identification and Control of Baker's Yeast Fermentation

We shall discuss in this section concepts on identification and control based in structured phenomenological models, with direct application to baker's yeast fermentation. The basic theory in described in detail by Bastin and Dochain (1990) and by Pomerleau and Perrier (1990) and Pomerleau and Viel (1992). The simulator described in Part I (Feyo de Azevedo *et al.*, 1992b) is employed for tests concerning the identification algorithms and their robustness to noisy measurements (Feyo de Azevedo *et al.*, 1992a).

3.1. PROCESS MODELLING

The dynamical model for the fed-batch fermentor is obtained from a mass balance on the components, considering that the reactor is well mixed, the yield coefficients are constant and the dynamics of the gas phase can be neglected. The kinetic model proposed by Sonnleitner and Käppeli (1986) is employed.

3.1.1. Kinetic model. Yeast growth is characterized by three metabolic pathways, viz-

$$S + C \xrightarrow{\mu_s^o} X + G \qquad \text{(respiratory growth on glucose)} \qquad (1)$$

$$S \xrightarrow{\mu_s^r} X + E + G \qquad \text{(fermentative growth on glucose)} \qquad (2)$$

$$E + C \xrightarrow{\mu_e^o} X + G \qquad \text{(respiratory growth on ethanol)} \qquad (3)$$

with S: glucose; C: oxygen; X: biomass; E: ethanol; G: CO_2 and $\mu^{o,s}$, $\mu^{r,s}$, $\mu^{o,e}$: specific growth rates for the three pathways. In the sequel X, S, E, C, G mean concentrations.

The metabolic pathways of oxidative growth on glucose and ethanol are governed by the respiratory capacity of the cells. If glucose is present in low concentrations and there is enough oxygen in the medium, only the oxidative metabolism takes place, being the glucose a preferable substrate rather than ethanol. On the other hand, if the glucose flow exceeds the respiratory bottleneck, part of it is catabolized oxidatively and the rest follows the fermentative catabolism, producing ethanol. In this case no oxidative growth on ethanol takes place.

The kinetics equations for baker´s yeast growth (equations (1)-(3)), considered as Monod equations, are determined as follows -

i. the total specific growth rate, μ_t, is the sum of the growth rates for the three pathways -

$$\mu_t = \mu_s^o + \mu_s^r + \mu_e^o \quad (4)$$

where μ_i can be related to the corresponding substrate fluxes, q, and yield coefficients, Y, viz -

$$\mu_t = - Y_{x/s}^o q_s^o - Y_{x/s}^r q_s^r - Y_{x/e}^{oe} q_e^o \quad (5)$$

ii. as ethanol uptake is influenced by the priority of glucose uptake, which functions as an inhibitor, the specific growth rate on ethanol can be described as -

$$\mu^{o,e} = \mu_{e,max} \frac{E}{E+K_e} \frac{K_i}{S+K_i} \quad (6)$$

where $\mu_{e,max}$ is the maximal specific growth rate, K_i is the inhibition parameter, K_e is the saturation parameter.

However, this equations holds true only if there is an available respiratory capacity of the cells.

iii. the glucose uptake, q_s,is sligthly different because it follows two metabolic pathways: oxidative and fermentative -

$$q_s = q_s^o + q_s^r \quad (7)$$

iv. the total flux may be described by the Monod kinetics equation -

$$q_s = q_{s,max} \frac{S}{S+K_S} \quad (8)$$

where $q_{s,max}$ is the maximal specific glucose uptake rate and K_s is the saturation parameter.

The oxidative glucose uptake depends on the availability of dissolved oxygen, and may be defined as -

$$q_s^o = \frac{q_{cs}}{a} \quad (9)$$

where a is the stoichiometric coefficient of the oxygen in the respiratory pathway of glucose and $q_{c,s}$ is the oxygen uptake on glucose. This last parameter also follows a Monod kinetics -

$$q_{c,s} = q_{c,s,max} \frac{C}{C+K_C} \quad (10)$$

with -

$$q_{c,s,max} = \min (q_{c,max}, aq_S) \quad (11)$$

being $q_{c,max}$ the maximal specific oxygen uptake rate.

3.1.2. Fermentor dynamic model. The mass balances, in terms of concentration, are written as -

$$\frac{dX}{dt} = (\mu_s^o + \mu_s^r + \mu_e^o - D) X \quad (12)$$

$$\frac{dS}{dt} = D (S_{in} - S) + (- \frac{\mu^{o,s}}{Y^{o,x/s}} - \frac{\mu_s^r}{Y_{x/s}^r}) X \quad (13)$$

$$\frac{dE}{dt} = - D E + (\frac{\mu_s^r}{Y_{x/e}^r} - \frac{\mu_e^o}{Y_{x/e}^{oe}}) X \quad (14)$$

$$\frac{dC}{dt} = - D C + OTR + (- \frac{\mu_s^o}{Y_{x/c}^o} - \frac{\mu_e^o}{Y_{x/c}^{oe}}) X \quad (15)$$

$$\frac{dG}{dt} = - D G - CTR + (\frac{\mu_s^o}{Y_{x/g}^o} + \frac{\mu_s^r}{Y_{x/g}^r} + \frac{\mu_e^o}{Y_{x/g}^{oe}}) X \quad (16)$$

and the additional equation -

$$\frac{dV}{dt} = F = D V \quad (17)$$

In the former - D is the dilution rate and the k_i (i=1 to 9) are the yield coefficients; S_{in} is the substrate concentration in the feed; OTR is the oxygen transfer rate (defined as OTR = $K_L a$ (C*-C) where $K_L a$ is the mass transfer coefficient and C* is the equilibrium concentration of dissolved oxygen) and CTR is the carbon dioxide transfer rate. The dynamics of CO_2 may be assumed as being very fast relatively to the dynamics of the other components. Also, the concentration of carbon dioxide at the pH of operation is known to be low. Hence the balance equation of CO_2 can be simplified by assuming dG/dt = 0 and G = 0. Defining Q_{CO2} as the gas outflow of CO_2, this leads to -

$$Q_{CO2} = CTR = (K_7 \mu_s^o + K_8 \mu_s^r + K_9 \mu_e^o) X \quad (18)$$

The set of equations above has a structure which can be represented by the general dynamic model proposed by Bastin and Dochain (1990) in the following matrix form:

$$\frac{d\xi}{dt} = K\varphi(\xi) - D\xi + F - Q(\xi) \quad (19)$$

where the notation emphasize that φ and Q may be time-varying and may depend on the process state ξ.

For the present case equations (12)-(16) take the matrix form:

$$\frac{d}{dt}\begin{pmatrix} X \\ S \\ E \\ C \\ G \end{pmatrix} = \begin{pmatrix} 1 & 1 & 1 \\ -k_1 & -k_2 & 0 \\ 0 & k_3 & -k_4 \\ -k_5 & 0 & -k_6 \\ k_7 & k_8 & k_9 \end{pmatrix} \begin{pmatrix} \mu_s^o \\ \mu_s^r \\ \mu_e^o \end{pmatrix} X - D \begin{pmatrix} X \\ S \\ E \\ C \\ G \end{pmatrix} + \begin{pmatrix} 0 \\ DS_{in} \\ 0 \\ OTR \\ 0 \end{pmatrix} - \begin{pmatrix} 0 \\ 0 \\ 0 \\ 0 \\ CTR \end{pmatrix} \quad (20)$$

The relevant kinetic data were taken from Sonnleitner and Käppeli (1986) (see table 1). The yield coefficients proposed by Pomerleau and Perrier (1990) were employed (see table 2).

Table 1. Kinetic parameters

Parameter	Value
$q_{s,max}$	3.5 g gluc g biom^{-1}h^{-1}
$q_{c,max}$	0.256 g O_2 g biom^{-1}h^{-1}
$\mu_{e,max}$	0.17 h^{-1}
K_e	0.1 gl^{-1}
K_i	0.1 gl^{-1}
K_S	0.2 gl^{-1}
K_C	0.1 mgl^{-1}

Table 2. Yield coefficient values

Coefficient	Value
$Y^{o}_{x/s}$	0.49 g biom g gluc^{-1}
$Y^{r}_{x/s}$	0.05 g biom g gluc^{-1}
$Y^{r}_{x/e}$	0.10 g biom g eth^{-1}
$Y^{oe}_{x/e}$	0.72 g biom g eth^{-1}
$Y^{o}_{x/c}$	1.20 g biom g O_2^{-1}
$Y^{oe}_{x/c}$	0.64 g biom g O_2^{-1}
$Y^{o}_{x/g}$	0.81 g biom g CO_2^{-1}
$Y^{r}_{x/g}$	0.11 g biom g CO_2^{-1}
$Y^{oe}_{x/g}$	1.11 g biom g CO_2^{-1}

3.1.3. Simulation with MIMOSA. Process simulation with MIMOSA (Feyo de Azevedo *et al.*, 1992b, Part I) is implemented by writing two files, *viz* - (i) BAKER1.B file where the right hand side of the model equations are defined (Table 3); (ii) BAKER1.D01 file - data file where variable and initial values are defined (Table 4).

Simulation is available in 'real-time', in 'scaled-time' and in 'simulation-time'.

During operation, loads to the process can be generated through the keyboard of the process computer. Also, the declared time-varying parameters (including delays and measurement noises) can be changed on-line and in real time. Finally, the appropriate control actions (if it is the case) generated by an external controller (computer or standard industrial system) can be sent to the process.

Figure 1 shows a run after 21.10 hours. In the upper half of the screen a graphics facility emulating a 4-pen register is available. In the lower half all main variables are monitored and made available for on-line change. In particular it should be noted that:

i. All system state variables y[ ] can be monitored and all are eligible as output variables with a superimposed 'user-defined' random noise;
ii. All parameters named as p[ ] in the model equations can be monitored and their values changed on-line. This includes the noises and dead-times defined in a data file, which MIMOSA automatically will add to the list of parameters and as such will make available for on-line change.

Table 3. BAKER.B file.

```
/*  Modelling baker's yeast.
    Universidade do Minho, Engenharia Biológica
    Gabinete de Sistemas (CEQ) do DEQ/FEUP
        PORTO    PORTUGAL, 92.01.07
    Eugénio Campos Ferreira
    Filomena Rocha Oliveira
    Pedro Pimenta                */

//  Checking ranges
if (y[1] <.0) y[1] = .0;       //[Substrate]
if (y[2] <.0) y[2] =     .0;       //[Ethanol]
if (y[3] > p[23])y[3]=p[23];   //[O2diss]<C*

//  Kinetic model.
p[25] = p[6] * y[1]/(p[9]+y[1]);
if ( (p[21] * p[25]) <= p[7])
        p[28] = p[25]*y[3]/(y[3]+p[26]);
else    p[28] = p[7]/p[21]*y[3]/(y[3]+p[26]);

p[1] = p[15] * p[28];
p[2]=p[8]*y[2]/(p[10]+y[2])*p[20]/(p[20]+y[1]);
p[3]=p[18]*( p[25]-p[28] );
p[5]=(p[1]/p[16]+p[2]/p[17])*y[0];
p[4]=(p[1]/p[13]+p[3]/p[14]+p[2]/p[12])* y[0];
p[0] = p[4]/p[5];                              //RQ

//Model of transient state
f[0]=(p[1]+p[2]+p[3]-c[0])*y[0];
f[1]=c[0]*(u[0]-y[1])-
    p[1]/p[15]+p[3]/p[18])*y[0];

f[2]=-c[0]*y[2]+(-p[2]/p[11]+p[3]/p[19])*y[0];
f[3]=-c[0]*y[3]+(p[22]*(p[23]-y[3]))-p[5];
f[4] = c[0] * y[4];

if( y[1] <= .0)                //[substrate]> 0.
{y[1] = .0; if( f[1] < .0) f[1] = .0;}

if( y[2] <= .0)                //[ethanol] > 0.
{y[2] = .0; if( f[2] < .0) f[2] = .0;}

if( y[3] <= .0)                //[oxygen] > 0.
{y[3] = .0; if( f[3] < .0) f[3] = .0;}

if( y[3]>=p[23])               //[O2] < C*
{y[3] = p[23]; if (f[3]>.0) f[3]=.0;}

f[0] = f[0] * p[30];           // Scaling in time...
f[1] = f[1] * p[30];
f[2] *= p[30];
f[3] *= p[30];
f[4] *= p[30];

// Stop conditions
if (y[4] >= p[27]){c[0]=f[4]=0.;p[29]=1.;}

if (p[29]==1.)             // Stationary state
{   p[24] = fabs(f[0])+
        fabs(f[1]) + fabs(f[2]) + fabs(f[3]);
    if (p[24] < 5e-5) fim(0, "STAT");
}
```

Table 4. BAKER.D01 file.

```
C  Initial Conditions for BAKER1
C  No. State variables
   5
C                Initial                Variancies   Delays
C  Name          Value  Min   Max      (noise)      (sec/100)
   Biomass       0.1    0.    3.       .1           0.
   Glucose       .8     0.    5.       .1
   Ethanol       .0     0.    2.       .1
   O2diss        .0066  0.    .007     5e-4         0.
   Volume        3.5
C  No. Loads
   1
C  Name          Initial Value
   Sin           10.
C  No. control actions
   1
C  Name          Starting Value
   D             .03
C  No. of parameters
   31
C                Initial
C  Name          Value     Comments
   RQ            1.0       p0
   µo            0.3       p1
   µe            0.        p2
   µr            .08       p3
   CER           0.0       p4
   OUR           0.0       p5
   qsmax         3.5       p6
   qO2max        .256      p7
   µemax         .17       p8
   Ks            .2        p9
   Ke            .1        p10
   Ye            .72       p11
   Yge           1.11      p12
   Ygo           .81       p13
   Ygr           .11       p14
   Yo            .49       p15
   Yo2           1.2       p16
   Yo2e          .64       p17
   Yr            .05       p18
   Yre           .1        p19
   Ki            .1        p20
   a             0.4142    p21
   Kla           100.      p22
   C*            0.007     p23
   SumDer        0.00      p24
   qs            0.0    m  p25
   Ko            .0001     p26
   Vmax          10.       p27
   qso           0.0       p28
   VMax?         .0        p29 - Flag for max. Volume
   TFact         .1        p30
```

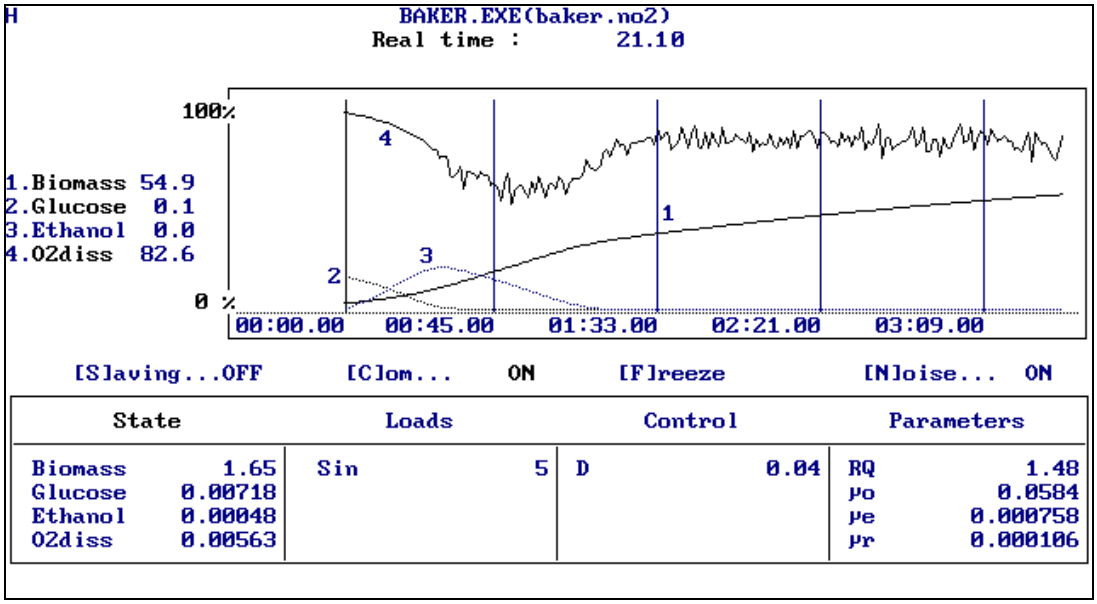

Fig. 1. Process simulation with MIMOSA.

3.2. THE OBSERVER ALGORITHM

We propose to develop an algorithm which will allow state and kinetic parameter estimation from a limited number of measurements, assuming that the yield coefficient are known.

Eq. (19) can be written as:

$$\frac{d\xi}{dt} = K\mu X - D\xi + U \tag{21}$$

in which: ξ is the vector of the bioprocess components (dim(ξ) = N); μ is the specific reaction rate vector (dim(μ)=M); *K* is the yield coefficients matrix (dim(K) = NxM); *U* is the feed and the gaseous outflow rates vector (dim(*U*) = N).

This state space representation can be divided in two partitions: the first one includes the equations related to the measured state variables (ξ_1); the second partition, the equations related to the nonmeasured state variables (ξ_2). The dynamic model is rewritten as:

$$\frac{d\xi_1}{dt} = K_1\mu X - D\xi_1 + U_1 \tag{22}$$

$$\frac{d\xi_2}{dt} = K_2\mu X - D\xi_2 + U_2 \tag{23}$$

where K_1 (a full rank matrix), K_2, U_1, U_2 correspond to the division of K and U according to each partition. A transformation Z is applied to the nonmeasured state variables partition, giving:

$$Z = \xi_2 - K_2 K_1^{-1} \xi_1 \tag{24}$$

$$\frac{dZ}{dt} = -DZ + U_2 - K_2 K_1^{-1} U_1 \tag{25}$$

A "Luenberger-type" asymptotic observer can be written using eq. (25) with Z replaced by its estimate $\hat{Z}$ and the non-measured state variables estimated by the following equation:

$$\hat{\xi}_2 = \hat{Z} + K_2 K_1^{-1} \xi_1 \tag{26}$$

For the specific growth rates the following estimator is employed:

$$\frac{d\hat{\psi}}{dt} = \hat{\mu} X - D\psi + K_1^{-1} U_1 + \omega(\psi - \hat{\psi}) X \tag{27}$$

$$\frac{d\hat{\mu}}{dt} = \gamma(\psi - \hat{\psi}) X \tag{28}$$

with $\psi = K_1^{-1} \xi_1$, a transformation to decouple the equations with respect to the specific growth rate; ω, γ are diagonal matrices containing time varying tuning parameters updated by a pole placement procedure (Pomerleau and Perrier, 1990). A discrete version of the estimator algorithms was implemented by first order Euler approximation with a sampling period of 6 minutes.

Two case studies illustrate the application. In both, the initial conditions were -

X(0) = 0.1 g/l, S(0) = 0.8 g/l, E(0) = 0 g/l, C(0) = 0.0066 g/l, V(0) = 3.5 l, $S_{in}(0)$ = 5 g/l.

The value for $K_L a$ was assumed as 60 hr^{-1}.

In the first a constant dilution rate of 0.04 hr^{-1} is assumed. The final volume of 10 L is taken as criterion to stop the feeding. Figure 1 represents the process simulation with MIMOSA.

Glucose, dissolved oxygen and ethanol were the measured variables available to the 'observer'. After about 3 hours a white noise of approximately 5% was imposed on the measurement of oxygen. The dotted lines in Fig. 3 represent the biomass and the flux of CO_2 estimated. The continuous lines represent the 'true' values obtained with the simulator. The reconstruction of the relevant specific growth rates is shown in Fig. 3. The influence of noise is visible in the figures, but it is well dumped by the filter.

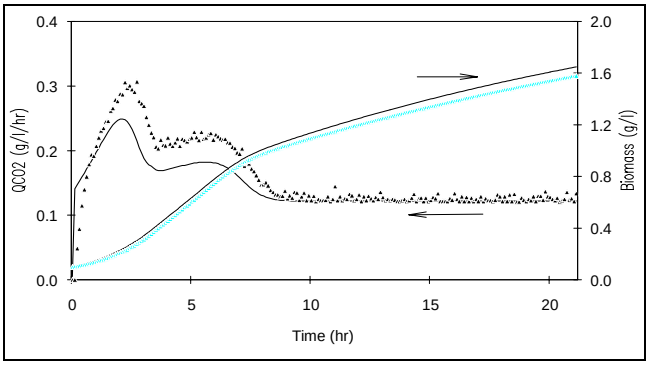

Fig. 2. 'Observed' - (dotted lines) vs. 'true' state properties (continuous line).

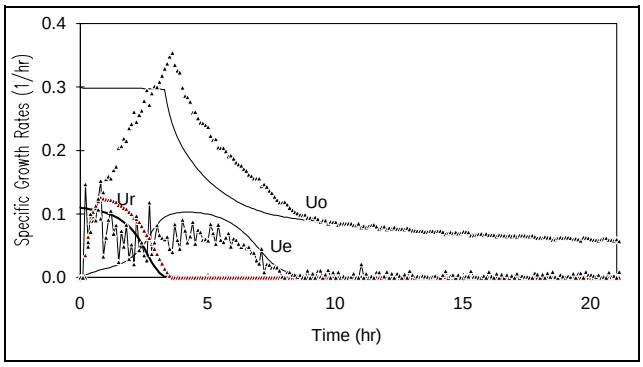

Fig. 3. 'Observed' - (dotted lines) vs. 'true' specific growth rates (continuous line).

In the second case study an exponential evolution of the dilution rate [D(t)=0.03 exp(0.1t)] was imposed in MIMOSA. A white noise with variance 0.01 was considered in the measurement of ethanol. Figure 4 shows the observed (dotted lines) and the 'true' values of biomass, glucose and CO_2 outflow obtained from single measurements of dissolved O_2 and ethanol. Here, a simplification had to be performed, having neglected the oxidative specific growth rate on ethanol, μ_e. The 'Observer' is not sensitive to the noise imposed and performs well.

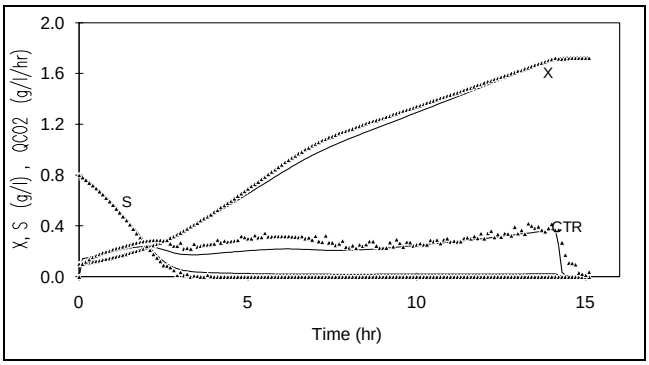

Fig. 4. Estimation of biomass, glucose and outflow of CO_2 from measurements of ethanol and dissolved oxygen.

3.3. EXACT LINEARIZING CONTROL

We shall discuss now the application of linearizing control to bioprocesses. The subject is proposed and extensively detailed by Bastin and Dochain (1990).

In what follows it will be seen that the non-linear structure of the model is fully exploited in solving the control problem. The other remark concerns the possible coupling of this strategy with an observer such as the one described in the previous section, in those cases where state variables are not directly available from measurements and/or parameters are unknown.

3.3.1. Brief theoretical description. The difference between this technique and conventional control lies in the way that linearization is introduced in the problem. In the standard approach, (like a PID control or MV control), we first calculates a linearized approximation of the model, and then we design a 'linear controller' for this approximate model. But the closed loop remains non linear. In exact linearizing control approach we obtain a 'non linear controller' which is precisely designed to achieve a 'linear closed loop'. This is summarised in Fig. 5.

Considering the fermentation process described by the general dynamic model (eq. (19)). The objective is to control a scalar output linear combination of the state variables:

$$y = \sum_{i=1}^{N} C_i \xi_i = C^T \xi \tag{29}$$

where $C^T = [C_1, C_2, ..., C_N]$ is a vector of known constants.

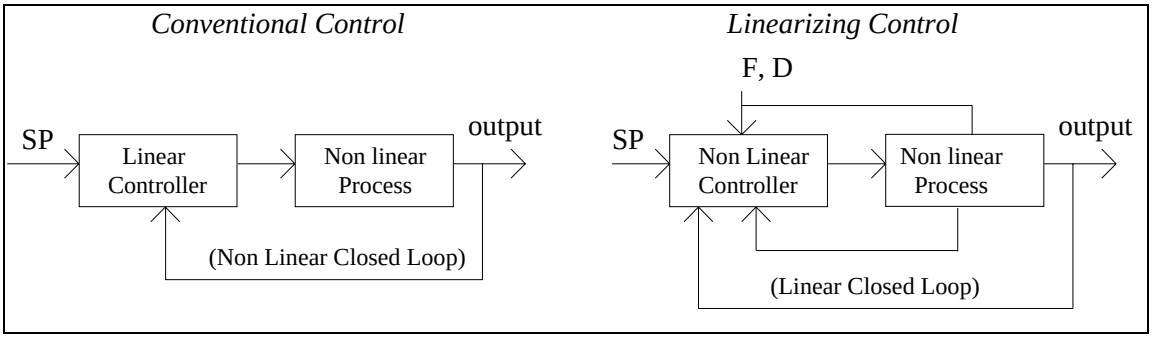

Fig. 5: Conventional Control vs. Linearizing Control

The control input (denoted by "u") is the feed rate of one substrate introduced to the process, from the outside.

$$u = F_i \quad \text{for some i} \tag{30a}$$

$$F = bu + f \tag{30b}$$

With this definition the model is rewritten as

$$\frac{d\xi}{dt} = K\varphi(\xi) - D\xi + bu + f - Q \tag{31}$$

It will be assumed that f and Q are measured on-line and that ξ is known on-line either by measurement or by an asymptotic observer.

The control objective is to track a reference output signal denoted y*(t) (set point is constant in a regulation problem).

The principle of linearization control is to find a control law u(ξ, Q, f, y*) which is a multivariable non linear function of ξ, Q, f e y* such that the tracking error e = (y*-y) is governed by a pre specified stable linear differential equation called a reference model.

Linearizing control design is a three-step procedure:

Step 1: to derive an Input/Output Model (I/O model) by successive differentiation of the general dynamic model:

$$\frac{d^{\delta}y}{dt^{\delta}} = f_0(t) + u(t)f_1(t) \tag{32}$$

where δ the order of the differential equation is called the relative degree.

Note that the I/O model is linear with respect to the control input u(t).

Step 2: A Stable Linear Reference Model of the tracking error is selected as follows:

$$\sum_{i=0}^{\delta} \lambda_{\delta-j} \frac{d^j}{dt^j}\left[y^*(t) - y(t)\right] = 0 \qquad (\lambda_0 = 1) \tag{33}$$

The coefficients $\lambda_{\delta-j}$ are chosen so that the differential equation is stable.

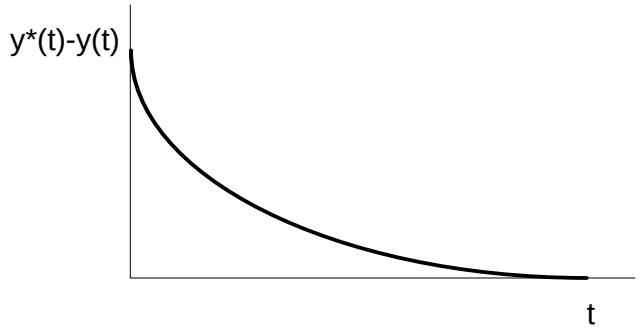

Fig. 6. Tracking error

Step 3: Control Design

To compute the control action u(t) such that the I/O model exactly matches the Reference Model

$$u(t) = \frac{1}{f_1(t)}\left[-f_o(t) + \sum_{j=0}^{\delta-1} \lambda_{\delta-j} \frac{d^j}{dt^j}\left[y*(t) - y(t)\right] + \frac{d^{\delta}y*}{dt^{\delta}}\right] \tag{34}$$

3.3.2. Application to Ethanol Regulation in a Yeast Fermentation Process. In the baker's yeast fermentation, the goal is to prevent the fed-batch reactor from ethanol (E) accumulation which is a source of yield decrease and may affect the productivity. Glucose feed rate is chosen as the control action.

$$y = E = C^T\xi \quad \text{with} \quad C^T = [0, 0, 1, 0, 0] \tag{35a}$$

$$u = F_1 = DS_{in} \quad \text{with} \quad b^T = [0, 1, 0, 0, 0] \text{ and } f^T = [0, 0, 0, OTR, 0] \tag{35b}$$

The control problem is solved by the 'Singular Perturbation Technique', which is justified by the following fact - *it is legitimate to assume that the kinetics of some substrates and products are much faster then some limiting ones*. The Singular Perturbation Technique is precisely a method to neglect fast kinetics and dynamics in ordinary differential equations in order to obtain a reduction of the dynamic model.

The following set of "fast" state variables is selected:

$$\xi_f^T = [S,\ C,\ G] \quad (M=3) \tag{36}$$

and for "slow" state variables

$$\xi_s^T = [X,\ E] \tag{37}$$

with $\xi^T = [\xi_s^T, \xi_f^T]$ (38)

It may be noticed that ethanol concentration E, which is the state variable to be regulated, does not belongs to ξ_f:

$$E = C_s^T\xi_s \quad \text{with} \quad C_s^T = [0,\ 1] \tag{39}$$

The general dynamic model is reduced to a set of M (=3) algebraic equations and N (=2) ordinary differential equations as follows:

$$\frac{d\xi_s}{dt} = K_s\varphi - D\xi_s + b_s u + f_s - Q_s \tag{40a}$$

$$K_f\varphi + b_f u + f_f - Q_f = 0 \tag{40b}$$

with

$$K_s = \begin{bmatrix} 1 & 1 & 1 \\ 0 & k_3 & -k_4 \end{bmatrix}, \quad b_s^T = [0, 0], \quad f_s^T = [0, 0], \quad Q_s^T = [0, 0] \tag{41a}$$

$$K_f = \begin{bmatrix} -k_1 & -k_2 & 0 \\ -k_5 & 0 & -k_6 \\ k_7 & k_8 & k_9 \end{bmatrix}, \quad b_f^T = [1, 0, 0], \quad f_f^T = [0, 0, 0], \quad Q_f^T = [0, \text{OTR}, -\text{CTR}]$$

(41b)

Since K_f is a full rank matrix, the vector $\varphi(\xi)$ of growth rates can be written as

$$\varphi(\xi) = K_f^{-1}(Q_f - F_f) = K_f^{-1}(Q_f - b_f u - f_f) \tag{42}$$

with the inverse of K_f computed as:

$$K_f^{-1} = \frac{1}{\tilde{k}} \begin{bmatrix} k_6 k_8 & k_2 k_9 & k_2 k_6 \\ (k_5 k_9 - k_6 k_7) & -k_1 k_9 & -k_1 k_6 \\ -k_5 k_8 & (k_1 k_8 - k_2 k_7) & -k_2 k_5 \end{bmatrix} \tag{43}$$

where $\tilde{k} \equiv k_2 k_6 k_7 - k_1 k_6 k_8 - k_2 k_5 k_9$. (44)

Substituting (42) into (40) gives

$$\frac{d\xi_s}{dt} = -D\xi_s + \left[I_{N-M} - K_s K_f^{-1}\right](F - Q) \tag{45}$$

The dynamics of Y (=E) is written as

$$\frac{dY}{dt} = -DY + C_s^T \left[I_{N-M} - K_s K_f^{-1}\right](F - Q) \tag{46}$$

or

$$\frac{dY}{dt} = -DY + C_s^T \left[I_{N-M} - K_s K_f^{-1}\right] bu + C_s^T \left[I_{N-M} - K_s K_f^{-1}\right](f - Q) \tag{47}$$

which leads to the following fully reduced input/output model:

$$\frac{dY}{dt} = -DY - \theta_1(CTR) - \theta_2(OTR) + \theta_3(DS_{in}) \tag{48a}$$

with

$$\theta_1 \equiv (k_1 k_3 k_6 - k_2 k_4 k_5)/\tilde{k} \tag{48b}$$

$$\theta_2 \equiv (k_2 k_4 k_7 - k_1 k_3 k_9 - k_1 k_4 k_8)/\tilde{k} \tag{48c}$$

$$\theta_3 \equiv (k_3 k_6 k_7 - k_3 k_5 k_9 - k_4 k_5 k_8)/\tilde{k} \tag{48d}$$

Selecting a first order reference model for the tracking error

$$\frac{d}{dt}(y^* - y) + \lambda_1(y^* - y) = 0 \tag{49}$$

the linearizing control law is readily obtained by substituting (22a) into (23) as follows

$$F_1(= DS_{in}) = \theta_3^{-1}\left\{\lambda_1(E^* - E) + DE + \theta_1(CTR) + \theta_2(OTR)\right\} \tag{50}$$

It is worth noting that the regulation law

i) does not need on-line measurement of the process compounds except that which is regulated (E);
ii) does not require the knowledge of the process kinetics;
iii) makes use of measurement of gaseous outflow rates which are, most often easily accessible on-line;
iv) involves a feedforward compensation of the influent substrate concentration S_{in} (if control action is the dilution rate).

If the yield coefficients which are present in θ_1, θ_2, θ_3 are either badly known or time-varying, an adaptive version of the controller must be implemented. The Adaptive Regulation law is obtained by using eq. (50) but with an on-line parameter estimation of θ_1, θ_2 and θ_3.

4. Closing Comments

Societies are periodically shaken by uncontrolled events which turn clear that 'quality and efficiency' must be in the first line of concern of any production policy, whatever other details are imposed by the market and social forces.

Twenty years ago the oil and energy crises cried for efficiency. Throughout the last decade individual demand and market competition pushed towards product diversity. The more recent collective concern for environmental and health problems is leading to policies of tight pollution control and monitoring of product specification. Labor costs keep increasing, predictably but steadily. The production policies for the next future will only be effectively and economically conducted through the use of computer-based technology and methods for process monitoring and control.

Today we have available tools and theories adapted to the practical application. We have efficient systems for process monitoring. We can implement improved PID-based algorithms and we reached the point where advanced model-based adaptive control algorithms can be used with robust industrial control systems. Other techniques, aiming at employing human experience in control decisions are actively being developed.

The motivation seems strong but we have to be fully aware that there are human, economical and technical difficulties in introducing new plant procedures, viz. -

Computer control systems are expensive and they generate significant running costs, particularly concerning specialised personnel. Also, there is a lack of trained engineers in this

field. This often results in a lack of internal motivation to promote changes and in inefficient use of systems already available in the plant. Another problem is that processes often change, leading to the need of modifying the software. Human resources have to be available. In all, there must be a decision on organizational changes to accomodate systems and applications engineers who will become part of the operating personnel.

The pace of changes is the net result of all these factors. But computers are definitely into plant operation.

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Acknowledgements - This work was partially supported by JNICT - *Junta Nacional de Investigação Científica e Tecnológica*, under contract numbers BD/224/90-IF and BD/1476/91-RM and INIC - *Instituto Nacional de Investigação Científica*.