Biodegradation **8:** 221–226, 1997.

Codigestion of olive oil mill wastewaters with manure, household waste or sewage sludge

I. Angelidaki & B.K. Ahring*
Department of Environmental Science and Technology, Building 115, The Technical University of Denmark, 2800 Lyngby, Denmark (author for correspondence)*

Accepted 24 June 1997

Key words: anaerobic digestion, olive oil mill effluents, manure, household waste, sewage sludge

Abstract

Combined anaerobic digestion of oil mill effluent (OME) together with manure, household waste (HHW) or sewage sludge was investigated. In batch experiments it was shown that OME could be degraded into biogas when codigested with manure. In codigestion with HHW or sewage sludge, OME dilution with water (1:5) was required in order to degrade it. Using continuously stirred lab-scale reactors it was shown that codigestion of OME with manure (50:50 and 75:25 OME to manure ratios) was successful with a theoretical OME utilization of 75% and with approx. 87% reduction of the lipids content in OME. An OME utilization of approx. 55%, and lipid reduction of 73% was reached in codigestion with HHW (50:50 and 75:25 OME to HHW ratios). The results showed that the high buffering capacity contained in manure, together with the content of several essential nutrients, make it possible to degrade OME without previous dilution, without addition of external alkalinity and without addition of external nitrogen source.

Introduction

Large amounts of organic waste are generated today from households, agriculture and industry. The major part of the waste from industry and households is deposited in landfills, an undesirable solution, because landfills disturb landscapes and leach pollutants into the groundwater. Furthermore, special wastes such as wastewaters from olive oil processing, are produced in large amounts in concentrated areas mainly in Mediterranean countries. Treatment of oil mill effluent (OME) still presents a problem, which has not been solved today. If not treated, this waste imposes a great environmental hazard, due to the very high organic COD loads.

Anaerobic digestion for treatment of OME possesses many advantages compared to aerobic treatment [6]. Anaerobic treatment of OME results in lower amount of sludge wastes, toxicants, and at the same time energy in the form of biogas is produced, which can be used for heating and electricity production. Several investigators have studied anaerobic degradation of OME and obtained COD-reductions up to approx. 80% [5–8, 17]. However, especially at high feed concentrations, the process proved to be unstable due to the inhibitory effect of polyphenols, lack of ammonia and due to the low alkalinity of OME. In order to overcome the above problems the OME was diluted with water, and urea was added as nitrogen supplement [7, 14]. Furthermore, the alkalinity of the reactors content is often adjusted by $NaHCO_3$, NaOH, or $Ca(OH)_2$ [7, 8]. However, dilution of OME with water results in unnecessary large effluent volumes, and addition of chemicals is not economical and environmentally desirable.

Codigestion is a waste treatment method where different types of wastes are treated together [2, 3]. Application of codigestion as an intelligent raw material management offers many advantages. The codigestion is expected to cost less than separate treatment systems, mainly due to the lower cost per volume treated at large plants. By dilution or concentration of the material to an appropriate dry matter content, for instance by mixing solid wastes with more diluted wastes such as liquid manure, a better handling and digestibility of the solid waste is achieved. Furthermore, dilution or counteraction of inhibitors such as ammonia or xenobiotic compounds is achieved as well certain nutrients are supplied to the waste [3]. It has been previously shown that codigestion of pig manure with ferrous containing waste resulted in decrease of H_2S formation [3]. Furthermore, detoxification of toxic compounds can be achieved by cometabolism which would be favourized by codigestion. A cometabolic process is defined as a microbial transformation of a compound by an organism which is unable to use it as energy or carbon source [16]. It has previously been shown that full dechlorination of pentachloro-phenols could only be achieved by codigestion with glucose [12]. OME contains polyphenol-like substances which are poorly degraded [6]. The possible positive effect of cometabolism could increase degradability of such compounds.

In the present work the effect of codigestion of OME together with manure, HHW and sewage sludge was examined.

Materials and methods

OME characterization

Fresh OME was obtained from an olive oil continuous centrifuge processing plant in Crete (Greece). The OME was gassed with nitrogen gas to avoid oxidation and placed in a refrigerated room (4 °C). The container with OME was gassed with nitrogen whenever opened in order to minimize autooxidation of the oil. Characteristics of OME are shown in Table 1.

Manure characterization

Cattle manure was obtained from the Lemvig biogas plant in Denmark. The manure was blended and was kept frozen (−20 °C) in 2 l portions. Characteristics of the cattle manure are shown in Table 1.

HHW characterization

The HHW was source sorted household waste obtained from the Helsingør biogas plant, containing mainly food and vegetable residuals. No garden waste was included. The waste was blended and diluted with water in order to result in the same VS content as for manure and was kept frozen (−20 °C) in 2 l portions. Characteristics of the HHW are shown in Table 1.

Batch experiments

In order to investigate the effect of codigestion of OME with manure, HHW or sewage sludge batch experiments were carried out in serum vials with a volume of 117 ml. Three series of batch digestions were prepared using digested manure, digested HHW or digested sewage sludge as inoculum. The vials were flushed with N_2:CO_2 (80:20) gas mixture to avoid entrance of oxygen, and 20 ml homogenized inoculum was anaerobically added. OME was added undiluted and in 1:2, 1:2.5 and 1:5 (OME:total-volume) dilutions with water at a volume of 20 ml. Controls without addition of OME were included, where OME addition was replaced with addition of 20 ml water.

The work was carried out using anaerobic techniques. The vials were placed in an incubator at a temperature of 55 °C, for the series with manure and HHW, and at 37 °C for the sewage sludge series corresponding to the operation temperatures in the reactors from where the inocula were taken.

CSTR experiments

Three 4.5 l CSTR reactors were used with a liquid volume of 2 l. The reactors were operated at a hydraulic retention time of 13 days at 55 °C.

Two reactors were fed with manure and the third reactor with HHW. After approx. 2 months of operation, OME was added to the feed of the two manure reactors at a concentration of 50:50 or 75:25 OME:manure ratios for the reactors $R_{M,50}$, and $R_{M,75}$ respectively. For the reactor fed with HHW, OME was added at a feed concentration of 50:50 OME:HHW ratio (reactor $R_{HHW,50}$), and after approx. two retention times the OME concentration was increased to 75:25 OME:HHW ratio ($R_{HHW,75}$).

OME utilization degree was calculated both for the batch and CSTR reactors. The OME utilization was calculated as the methane produced from the reactor or the vial minus the experimentally determined methane potential of the manure, HHW or sewage sludge divided by the theoretical methane potential (based on Buswell's formula) of the OME fed to the reactors. The methane potential of manure, HHW and sludge were determined from the controls where OME was not added. The OME utilization calculations were based on the assumption that all the carbon is transformed to biogas. In reality some of the carbon will be consumed for cell production, and other maintenance losses, resulting in an overestimation of the theoreti-

Table 1. Characteristics of the wastes used

	Unit	OME	Manure	HHW
TS	g/l	60.3 (1.4)	62.6 (1.1)	50.7 (0.9)
VS	g/l	51.3 (0.8)	46.0 (0.9)	46.0 (0.8)
COD	g/l	104.9 (1.6)	103.4 (6.7)	103.5 (1.8)
Ash	g/l	8.9 (4.6)	16.7 (2.0)	5.0 (0.9)
VFA	g/l	1.5 (0.1)	4.8 (0.2)	0.5 (0.0)
Ammonia-N	g-N/l	0.1 (0.0)	2.5 (0.1)	0.47 (0.0)
Total-N	g-N/l	1.2 (0.0)	3.5 (0.1)	1.2
pH		4.81	7.15	4.10
Alkalinity	mM-H+	55	282	n.d.
Proteins	g/l	6.9	6.3	4.6
Lipids	g/l	17.2	2.5	5.3

Numbers in parentheses are standard deviations. TS: total solids; VS: volatile solids; COD: Oxygen Chemical Demand; n.d. non determined; VFA: volatile fatty acids.

cal methane potential of the OME and thus an underestimation of the OME utilization degree of approx. 5–10%.

Analytical methods

Volatile solids, total solids, chemical oxygen demand and pH were determined using standard methods [10]. CH_4 and CO_2 content of biogas produced from reactors in the continuous experiments were determined with gas chromatography using TCD detection as previously described [4]. VFA were analyzed on a gas chromatograph equipped with an FID detector [4]. Ammonia content was determined using the Kjeldahl method (steam distillation of ammonia). Lipids were determined by the Soxlet method [10].

Results and discussion

Batch experiments

OME could be degraded to biogas with a degradation degree of 77%. This degree of degradation is in accordance with previously reported values in the range of 60 to 80% found as COD reductions during anaerobic degradation of diluted OME [17]. OME could be degraded without dilution by codigestion with manure. However, when OME was added without dilution the process was slightly inhibited compared to the vials where OME was diluted with water (1:2 to 1:5 OME:total-volume) which reached an OME utilization of 77% (Figure 1a). In codigestion with HHW approx.

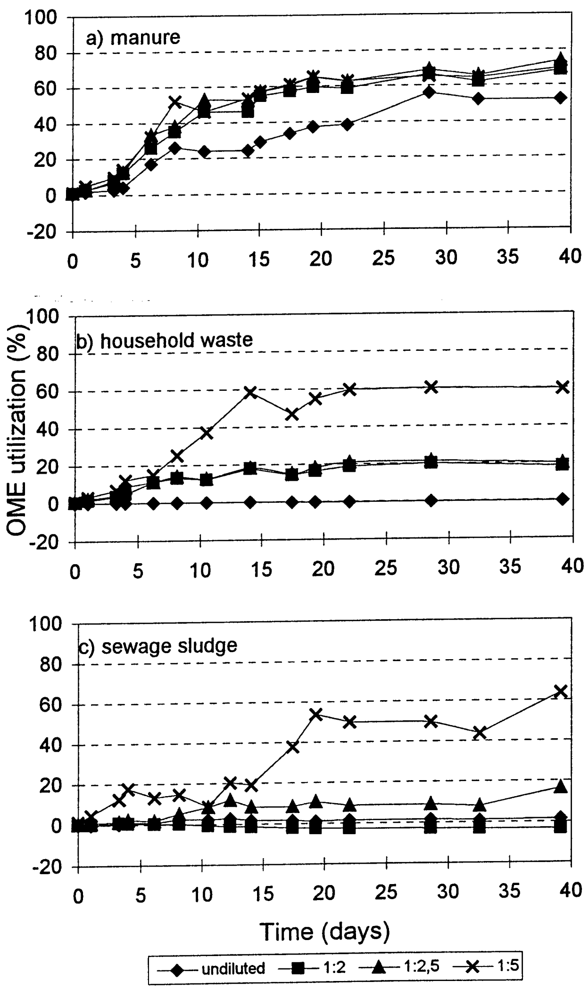

Figure 1. OME utilization a) with manure b) with HHW and c) with sewage sludge. Symbols: ♦: undiluted; ■: 1:2; △: 1:2.5 and x:1:5 dilutions of OME in water.

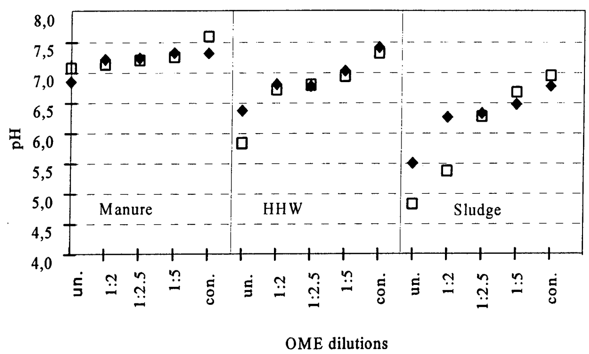

Figure 2. pH in batch experiments inoculated with: a) with manure b) with HHW and c) with sewage sludge. Symbols: ♦: at the start of the experiment; □: at the end of the experiment. (X-axis represents dilutions of OME in water; un represents vials with undiluted OME addition and con: represents control vials with no OME addition).

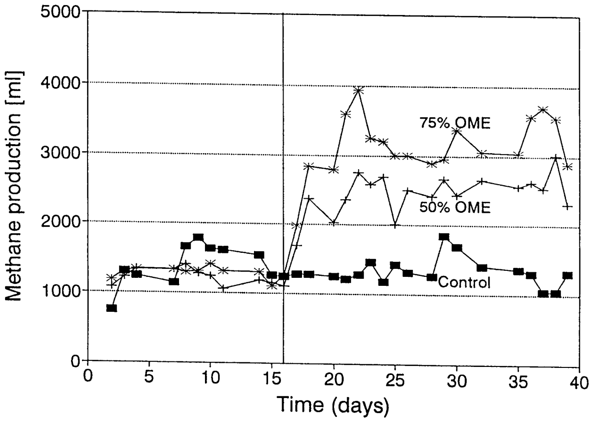

Figure 3. Methane production of the CSTR reactors fed with manure and OME. The vertical line marks when the feed was changed from manure alone to: ■ : Control (fed with manure only); +: $R_{M,50}$ (fed with 50:50 OME:Manure); ✳: $R_{M,50}$ (fed with 75:25 OME:Manure).

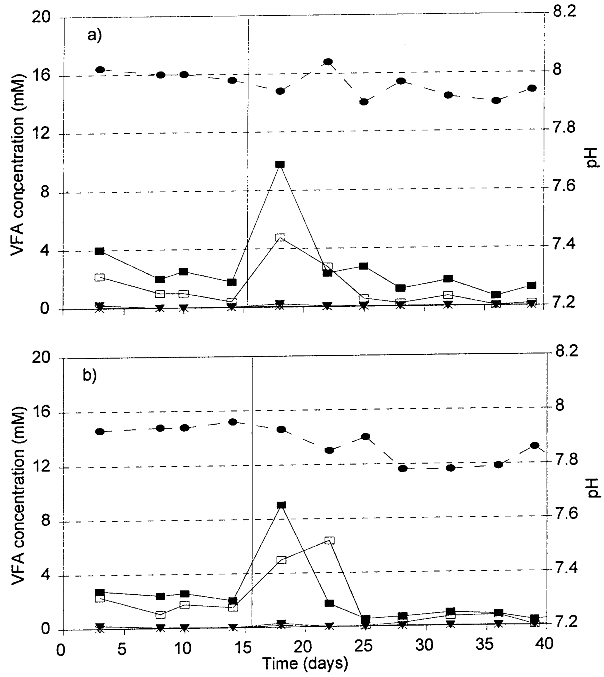

Figure 4. VFA concentrations and pH in the CSTR reactors fed with manure and OME. The vertical line marks when the feed was changed from manure alone to OME:manure feed. a) $R_{M,50}$ (fed with 50:50 OME:Manure); b) $R_{M,75}$ (fed with 75:25 OME:Manure). Symbols: ●, pH; ■ , acetate; □ , propionate; △, isobutyrate; ▲, butyrate; x, valerate.

60% utilization of OME could be achieved only in the highest dilution (1:5) (Figure 1b). Lower dilutions (1:2 and 1:2.5) resulted in only 20% OME utilization and the undiluted OME resulted in complete inhibition of the biogas process (Figure 1b). When codigested with sewage sludge a satisfactory degradation of OME could only be obtained at a high dilution (1:5) (65% OME utilization) (Figure 1c). All lower dilutions resulted in serious inhibition of the biogas process.

Measurements of pH in the vials showed that the vials which were strongly inhibited also had the lowest pH (Figure 2). OME addition, resulted in decrease of the pH, in the vials with HHW and sewage sludge. Addition of undiluted OME to the vials inoculated with digested sewage sludge resulted in a drop in pH down to 5.0 which is lower than the pH allowing growth for methanogenic bacteria. Methanogens have a limited pH span ranging from approx. 6.0 to 8.0 [15]. The strong buffering capacity of manure resulted in a pH in the range from 7 to 7.5 in vials inoculated with digested manure (Figure 2).

A drop in pH at the end of the experiment was further measured in the vials inoculated with digested sewage sludge, with undiluted and 1:2 diluted OME additions (Figure 2). This indicates that the acidogenic activity still occurs under these conditions, resulting in VFA production and a decrease in the pH. This is in accordance with the fact that most methanogens have pH optimum in the range 7 to 8, while acidogens have a lower pH optimum [15].

The results show that manure has a positive effect on OME degradation. Manure possesses a high alkalinity 282 mM-H^+, while the alkalinity in OME was 55 mM-H^+ (Table 1). Combining manure with OME will result in an alkalinity of approx. 169 or 131 mm-H^+ for 50:50 and 75:25 OME:manure combinations, respectively. Thus, codigestion of OME with manure results in increased resistance to acidification.

Manure, has a relatively high content of ammonia (2.5 g-N/l), while OME has a very low of ammonia content (0.1 g-N/l) (Table 1). Bacteria require nitrogen for cell growth and nitrogen requirement in a reactor will be correlated to the bacterial biomass synthesized, and thus the organic loading of the reactor. Reactors operated with addition of OME have a higher biogas production and bacterial activity than the reactors operated with manure alone, and thus, will have a higher need for nitrogen. Ammonia (NH_4^+ and NH_3) is an important component for bacterial growth as nitrogen source in a reactor environment. Concentrations between 50 and 200 mg/l are shown to be beneficial for bacterial growth [13]. Many ruminal bacteria, and the

Table 2. Results from the CSTR experiment

Reactor	COD loading (g-COD/ld)	OME-COD loading (g-COD/ld)	Theoretical[1] OME utiliz. (%)	Practical[2] OME utili (%)	Reduc. in lipids (%)	VFA (g/L)
$R_{M,50}$	7.8	3.9	75	106	85	0.13
$R_{M,75}$	7.8	5.9	70	102	87	0.09
$R_{HHW,50}$	7.5	3.9	55	83	73	0.11
$R_{HHW,75}$	7.7	5.9	57	85	n.d.	0.38

n.d. non determined; [1] Theoretical OME utilization is calculated as the biogas produced from the reactor minus the biogas potential in the corresponding manure or HHW divided by the theoretical biogas potential in the OME; [2] Practical OME utilization is calculated as the biogas produced from the reactor minus the biogas potential in the corresponding manure or HHW divided by the highest biogas potential reached in the batch experiments.

cellulolytic bacteria in particular, utilize ammonia-N in preference to amino acid-N; ammonia is an absolute growth requirement for some species [9]. A correlation between free-ammonia concentration and the concentration of volatile fatty acids in a biogas reactor showed a sharp increase of the VFA for free-ammonia concentrations lower than 0.01 g-N/l, indicating that ammonia can be limiting for bacterial growth [11]. Codigestion with even small amounts of manure eliminate the risk of ammonia deficiency.

The initial COD-concentration for the undiluted OME vials was quite high (52 g-COD/l) compared to previously reported limits for stable digestion. Boari et al. [7] reported that the maximum concentrations of OME should be around 18 g-COD/l when digesting OME in an upflow anaerobic sludge blanket reactor.

A biogas potential of approx. 40 l-biogas/kg-OME was determined from the 1:5 OME dilution codigested with manure.

CSTR experiments

The methane production increased from approx. 1200 ml/day to approx. 2500 ml/day or 3100 ml/day for the reactor $R_{M,50}$ and $R_{M,75}$ when the reactor feed was changed from manure alone to manure: OME mixture although manure and OME had the same VS and COD content (Figure 3). The reason for the increase can be attributed to both the higher biodegradability of OME compared to manure which typically contains high amounts of scarcely degradable lignocellulytic matter, and to the higher content of lipids of OME compared to manure (Table 1). When household waste was changed to combinations of OME and household waste, no significant increase of the methane production was observed, due to the same quality and strength of the organic matter in household waste and in OME (data not shown).

After introduction of OME into the reactors VFA increased initially. However, after approx. 10 days VFA concentrations returned to the level as before OME addition (Figure 4). The low level of VFA (Table 2) indicates that the process was stable. After addition of OME in reactors, $R_{M,50}$ and $R_{M,75,}$ only a slight decrease of pH was observed and the pH stabilized to approx. 7.9 and 7.8 for reactor $R_{M,50}$ and $R_{M,75}$, respectively (Figure 4). The pH in the reactors fed with HHW was approx. 7.2, with a slight decrease to 7.05 when 75% OME was added (data not shown).

Reactor $R_{M,75}$ stabilized at a lower VFA level after introduction of OME compared to the level before OME addition (Figure 4b), indicating that the increased bacterial activity and/or the lower concentration of ammonia, have resulted in increased reactor stability. This is in accordance with previous findings where higher loading resulted in increased stability of the process [1].

OME could successfully be degraded in codigestion with manure and HHW (Table 2). Especially when codigested with manure the OME utilization degree was high, reaching values of up to 75%. Comparing the biogas production originating from OME in the CSTR reactors, to the experimentally determined biogas potential of OME from the batch experiments, a practical OME utilization was determined. It was shown that in reactors $R_{M,50}$ and $R_{M,75}$, OME had a practical utilization of 106 and 102%, respectively, indicating that in these reactors all the biogas potential of OME was obtained (Table 2). Lower values of practical OME utillization (approx. 85%) were found for $R_{HHW,50}$ and $R_{HHW,75}$ (Table 2).

OME-utilization degrees of approx. 55% were achieved during HHW-OME codigestions (Table 2).

These results are consistent with the batch experiments, where the best performance was observed for manure-OME codigestions. The lower OME utilization degrees achieved for HHW-OME codigestions compared to manure-OME codigestions can be explained by the lower ammonia levels of HHW compared to the ammonia levels in manure.

Combined treatment of manure together with industrial waste and household waste produce both biogas and a fertilizer which can be used on farmland. At the same time manure is an abundant source which often will be available for codigesting of wastes such as olive oil mill wastewaters which are difficult to digest alone.

Acknowledgements

We thank Claudio Albano, Jacub Horecký and Hector Garcia for their help during this work. This work was supported by grants from the research programme of The Danish Energy Council, no, 1383/93–2

References

1. Ahring BK (1995) Methanogenesis in thermophilic biogas reactors. Ant. van Leeuw. 67: 91–102
2. Ahring BK, Angelidaki I & Johansen K (1992) Anaerobic treatment of manure together with organic industrial waste. Water Sci. Technol. 7: 311–318
3. Ahring BK, Angelidaki I & Johansen K (1992) Co-digestion of organic solid waste, manure and organic industrial waste. Waste Management International K.J. Thimé-Kozmiensky (Eds.) EF-Verlag für Energie-und Umwelttechnik GmbH 1: 661–666
4. Angelidaki I, Petersen S & Ahring BK (1990) Effect of lipids on thermophilic anaerobic digestion and reduction of lipid inhibition upon addition of bentonite. Appl. Microbiol. Biotechnol. 33: 469–472
5. Aveni A (1984) Biogas recovery from olive oil mill wastewater by anaerobic digestion. In: Rreffero GL, Ferranti MP & Naveau H (Eds) Anaerobic digestion and carbohydrate hydrolysis of waste (pp 489–491). Elsevier Applied Science Publishers, Essex
6. Beccari M, Bonemazzi F, Majone M & Riccardi C (1996) Interactions between acidogenesis and methanogenesis in the anaerobic treatment of olive oil mill effluents. Wat. Res. 1: 183–189
7. Boari G, Brunetti A, Passino R & Rozzi A (1984) Anaerobic digestion of olive oil mill wastewaters. Agricul. Wastes 10: 161–175
8. Boari G, Mancini IM & Trulli E (1984) Anaerobic digestion of olive oil mill effluent pretreated and stored in household waste sanitary landfills. Wat. Sci. Technol. 28: 27–34
9. Bryant MP & Robinson IM (1961) Studies on nitrogen requirements of some ruminal cellulolytic bacteria. Appl. Microbiol. 9: 96–103
10. Clesceri LS, Greenberg AE & Trussel RR (1985) Standard methods for the examination of water and wastewater. American Public Health Association, Washington, D.C.
11. Hashimoto AG (1983) Conversion of straw-manure mixtures to methane at mesophilic and thermophilic temperatures. Biotechnol. Bioeng. 15: 185–200
12. Hendriksen HV, Larsen S & Ahring BK (1992) Influence of supplemental carbon source on anaerobic dechlorination of pentachlorophenol in granular sludge. Appl. Environ. Microbiol. 58: 365–370
13. McCarty, 1964. Anaerobic waste treatment fundamentals III. Public works 95: 91–94
14. Morelli A, Rindone B, Andreoni V, Villa M, Sordini C & Balice V (1990) Fatty acids monitoring in the anaerobic depuration of olive mill wastewaters. Biological Wast. 32: 253–263
15. Sørensen AH (1996) Microbial characterization of methanogenic reactors. Ph. D. Thesis. Copenhagen, Denmark
16. Schink B (1988) Principles and limits of anaerobic degradation: Environmental and technological aspects. In: Zehnder AJB (Ed) Biology of anaerobic microorganisms. Chapter 14. A Wiley-Interscience Publication. John Wiley & Sons.
17. Tsonis SP & Grigoropoulos SG (1993) Anaerobic treatability of olive oil mill wastewater. Wat. Sci. Tech. 28: 34–44